

# PROJECT FINAL REPORT

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## 1. Final publishable summary report

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Serotonin and GABA are two neurotransmitters that have key roles in the control of anxiety states as they are main targets of anxiolytic drugs. However the neurobiological mechanisms of their action are still not well understood. The rationale of the project was based on new findings brought about by members of this consortium that radically change our views of the neurobiological action of these neurotransmitters. The first original finding was the discovery of a developmental role of 5-HT in the genesis of anxiety disorders, and the finding of interactions between 5-HT-related genes and environmental risk factors. The second finding was the discovery that metabotropic GABA-B receptors play a critical role in mediating the anxiolytic effects of GABA. Finally, evidence pointed to strong reciprocal interactions between the two systems: 5-HT influences the GABAergic regulation of emotion and GABA-B and 5-HT<sub>1A</sub> receptors converge on common signaling pathways.

The seven teams that composed the consortium had complementary expertise in development, neural plasticity, neurobehavior, pharmacology, and advanced mouse genetics. They joined forces to elucidate the neural circuits that are controlled by 5-HT and GABA-B receptors and to investigate their interaction in neural structures that are known to be involved in the control of fear and anxiety, namely the raphe, the hippocampus and the amygdala.

The research lead in the consortium allowed the following significant advances for the field:

- Characterization of new models of 5-HT depletion and identification of the effects of 5-HT depletion at different developmental times.
- Obtaining new tools for targeting the 5-HT raphe neurons.
- Revealing a genetic heterogeneity in the raphe serotonin neurons.
- Identifying with pharmacogenetic silencing tools circuits that are involved in anxiety responses (contextual fear conditioning).
- Identifying the brain regions specific for 5-HT<sub>1A</sub> developmental programming of anxiety.
- Characterizing the interactions between serotonin and GABA-B receptors systems in the hippocampal circuits and their roles in adult neurogenesis.
- Identifying the plasticity mechanism in hippocampal neural circuits involved in associative learning conducting to fear.
- Discovering new pathways of GABA-B signaling: the KCTD proteins, as modulators of GABA-B receptors.
- Characterizing the cellular and developmental impacts of GABA-B signaling for adult anxiety behaviors.
- Identifying new interactions between genes and the environment in the control of adult anxiety behaviors.
- Identifying the role of microbiota in the control of anxiety disorder.

These discoveries indicate the way to new paths for drug discovery or behavioral therapy in the field of anxiety disorders, which constitute a major societal burden.

## 1.1 Executive Summary

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### **The part played by serotonin in anxiety control**

In patients suffering from depression, panic attacks, anxiety disorders or phobias, the administration of drugs that increase serotonin (5-hydroxytryptamine, 5-HT) transmission has beneficial effects. However, the underlying mechanisms are still not elucidated, and a causal role for 5-HT depletion in anxiety disorders is not established.

The difficulty arises from the fact that 5-HT is produced in several organs besides the brain, and that it affects most all brain areas with sometimes opposing effects. Moreover 5-HT has clear developmental effects so it is unclear whether effects on anxiety reflect changes in neural circuit wiring or direct effects of 5-HT depletion. To answer these questions, researchers in the DEVANX consortium decided to characterize animal models with 5-HT depletion only in the brain, that are either complete or limited to some brain regions. To determine whether changes are developmental, they used rescue strategies, or induced 5-HT depletion in adults. Different mouse models were used relying on different mechanism: 1) Mouse strains with natural variation in the activity of the enzyme controlling central serotonin synthesis, TPH2 (BalbC/C57BL/6). 2) Pet1-KO mice in which the production of a subpopulation of raphe neurons in the brain is arrested during development by the lack of a transcription factor (Pet1-KO). 3) Conditional VMAT2-KO mice in which the vesicular storage of amines is prevented specifically in the serotonin raphe neurons (VMAT2-Sert-Cre; VMAT2-Pet1-Cre;). 4) Dietary restriction of the 5-HT precursor tryptophan; 5) Pharmacogenic silencing of the firing of raphe neurons.

The comparison of these different models (by Partners, 1, 2, 4 and 6) taught us that severe serotonin depletion, whatever the cause, had dual effect on anxiety related behaviors, depending on the type of behaviors that are explored. On the one hand serotonin depletion increased learned fear and escape responses. On the other hand serotonin depletion had anxiolytic effects in tests measuring exploration (Elevated plus maze) or conflict tests (Novelty suppressed feeding). These effects did not appear to be developmental, since they could be rescued by clorgyline in VMAT2 conditional KO mice or reproduced by dietary restriction of tryptophan. These results may best be explained in the general framework of involvement of 5-HT in behavioral inhibition, with different outcomes according to the behavioral test/ brain regions that is controlled by 5-HT.

Interestingly these studies also showed that the reduced 5-HT synthesis that is observed in the Balb/c mouse strain, due to a mutation of the TPH2 gene, does not explain their increased anxiety phenotype which could rather be due to adaptive changes in the serotonin raphe system with functional desensitization of the 5-HT<sub>1A</sub> receptors, causing an increased excitability of these neurons.

Another important finding from these studies was the existence of heterogeneity of central serotonin systems, and the identification of raphe neurons genetically defined by their lack of requirement for the Pet1-transcription factor and a selective innervation of brain areas involved in stress response.

In parallel Partner 6 obtained new promising models to control selectively gene expression within the serotonin raphe neurons. The new mouse line with selective

drivers (Tet- on and Cre-Ert2) that were generated will allow to drive or repress expression of genes selectively in the raphe with a tight temporal control.

Finally on a circuit level, the targets of 5-HT innervation that are important in mediating the developmental impact of 5-HT<sub>1A</sub> receptor invalidation were clarified (Partner 2). It had been previously found that developmental ablation of 5-HT<sub>1A</sub> receptors in the forebrain was sufficient to cause anxiety phenotypes. During this project researchers demonstrated that selective rescue of 5-HT<sub>1A</sub> in more selected regions of the forebrain was sufficient to restore normal responses to anxiogenic situations. This Partner also identified cellular effects of the 5-HT<sub>1A</sub> receptors on dendrite maturation in the hippocampus.

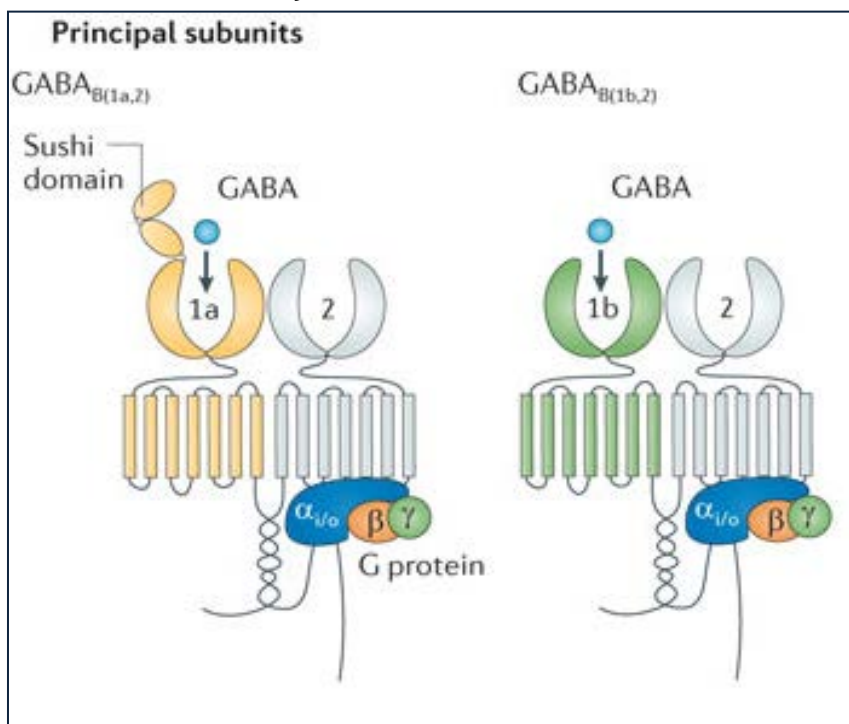
## **The GABA-B receptors**

Dysfunction of the gamma-amino butyric acid (GABA)-ergic system has been purported to play a role in anxiety and depression. A clear link between GABA-A receptors and anxiety has long been established since most anxiolytics such as benzodiazepines are positive allosteric modulators of GABA-A receptors. However the functional role of another major signaling pathway for GABA, the GABA-B receptors, has been less explored. GABA-B receptors are expressed largely in the brain, are targeted by new molecules that work in a completely different way from conventional anxiolytics. Partners 3 and 4 of this consortium were involved in the development of a novel class of GABA-B receptor allosteric modulators, and developed a number of genetic tools to understand GABA-B receptor function. Within the framework of this project a better understanding of the structure and the function of GABA-B receptors was achieved and their interactions with the serotonergic system, were clarified.

Partner 3 demonstrated that GABA-B receptors are heterodimers (a combination of two different receptor subunits) that possess associate proteins capable of modifying their binding properties. The pharmacological properties of GABA-B receptors vary depending on how the combination of subunits and associate proteins are organized. Partner 3 identified new auxiliary proteins for GABA-B receptors that contribute to the functionality of the receptors. Partner 4 showed that inhibiting GABA-B receptors can reduce depressive behavior in adults and has an effect on adult neurogenesis; they moreover were able to demonstrate a moderate anxiolytic effect of the GABA-B positive modulator. Partner 1 studied the connection between GABA-B receptors and the serotonergic system. Finally Partner 2 showed that interfering with GABA transmission (with benzodiazepines on the GABA-A receptors) caused long term effects on anxiety-related behaviors.

## GABA-B receptor subunits: GABA-B1a, GABA-B1b and GABA-B2.

(Copyright Gassmann and Bettler, 2012)



The principal subunits of GABA-B receptors have the prototypical seven transmembrane domains of G-protein coupled receptors and form two distinct core units: GABA-B (1a, 2) and GABA-B (1b, 2). Whereas the GABA-B1 subunits contain the GABA binding site, GABA-B2 subunits couple to the G protein. GABA-B1a and GABA-B1b are subunit isoforms that differ by the presence of two amino-terminal sushi domains in GABA-B1a.

## Other neural circuits involved in fear control

It is becoming more and more evident that it is the normal neuron circuits specialized in dealing with fear that are pathologically affected or amplified in anxiety disorders. Thus it is very important to understand and analyze how these circuits function in "real situations" in animal models. The end purpose is to find a way of "deconditioning" certain brain circuits that have been abnormally or over-activated.

The new approaches to physiology on the knockout animal, combined with pharmacogenetic research, have made for progress in this field. For example Partner 5 recorded different neurons from the hippocampal circuits in different fear learning situations and observed the effect of modifying the message conveyed by GABA-B and serotonin. Partner 2 used the serotonergic receptors (5-HT<sub>1A</sub>) expressed in different areas of the brain in order to temporarily deactivate highly specific neuronal circuits. This allowed them to identify the hippocampal and amygdala circuits involved in the generalization of fear.

Partner 4 identified the role of the rostral anterior cingulate cortex in modulating the efficiency of amygdala dependent fear learning, and demonstrated antidepressant effects of transient inactivation of the prefrontal cortex.

Partner 1 identified synaptic specializations of 5-HT axons in certain parts of the brain.

In parallel Partner 5 also identified the medial prefrontal cortex as having a key role in observational learning which may play significant roles in anxiety-related disorders.



## Gene-Environment interactions

Risk for anxiety-related disorders is determined by a combination of genetic and environmental factors. In particular traumatic childhood experience such as maternal separation can play a role in modifying the individual's response to stress in adult life. This effect can be modulated by specific polymorphisms in genes that control neurotransmission.

Researcher in the Devanx consortium identified several important factors in this regard. Partner 1 analyzed epigenetic changes in *bdnf* gene which occurred during anxiety-depressive disorders.

Partner 2 showed with a genetic approach that the serotonin transporter (5-HTT) and neurotrophic factor, BDNF, both moderate the effects of early maternal separation: when one allele of these genes is absent the effects of maternal separation on adult anxiety behaviors such as ambiguous fear conditioning.

Partner 4 demonstrated the influence of early life stress of the brain-gut axis disorders. They showed that stress occurring early in life alters brain-gut axis function and modifies the relative diversity of the gut microbiota, causing visceral hypersensitivity. They also showed a major effect of the genetic background on the resilience to early life stressors, as quite strikingly in some genetic backgrounds, paradoxical anxiolytic effects were observed after early prolonged maternal separation. Finally they also showed that there is an interaction between the GABA-B system and early life stress in terms of susceptibility to the effects of stress at a behavioral level and in the context of hippocampal neurogenesis.

## Conclusion

Research of this consortium into anxiety disorders, combined several fields of neurosciences, combining molecular approaches with integrated approaches in live animal. The genetic tools used or produced in this project provide unequalled power for researching into a determined molecule function, or a molecular assembly within a given circuit and a precise time slot. This type of approach will continue to develop in the years to come, with the coming of tools that will allow us to activate or deactivate certain selected neuron circuits.

By solving these intertwined elementary processes step by step, and going from molecular approaches to integrated physiological and behavioural approaches, in animal models clear progress can be made in the explanation to the mechanisms underlying pathological anxiety disorders.

## 1.2 Summary description of project context and objectives

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The project focuses on elucidating the mechanisms by which GABAergic and serotonergic systems act in the developmental programming of anxiety. New findings, brought about by members of this consortium, changed our views on the neurobiological action of these two transmitters and are the focus of the present proposal. The first original dimension is the discovery of a developmental role of serotonin (5-HT) in the genesis of anxiety disorders, and the finding of interactions between 5-HT-related genes and environmental risk factors. The second new dimension is the discovery that metabotropic GABA-B receptors play a critical role in mediating the anxiolytic effects of GABA, a starting point for the conception and design of novel therapeutic approaches.

Researchers that are at the forefront of these research domains will build on and extend new findings that involve the production of new genetic models in mice for site- and time-specific invalidation of 5-HT related genes, focusing on the hippocampus, amygdala and raphe nuclei. They will explore the role of GABA-B receptors in anxiety and the interaction of these receptors with the 5-HT system. The developmental effects of GABA-B receptors will be explored and a new generation of GABA-B modulators that produce anxiolytic effects in animal models. Finally they will investigate how exposure to adverse environments interacts with 5-HT-genes and GABA-B receptor genes to produce anxiety phenotypes.



### 1.3 Description of the main S&T results/foregrounds

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#### ***WP-1-Producing/characterizing mice with different serotonin depletion***

While supported by large circumstantial evidence, the causal role of 5-HT dysregulation in anxiety disorders is not yet established. It remained to be shown: 1) that the serotonergic dysfunction in anxiety is causal and not an adaptive symptom, 2) that 5-HT is deregulated in the brain rather than in the periphery, 3) that the timing of 5-HT deregulation during different life time periods plays different causal roles to generate anxiety, and 4) that 5-HT innervation to different brain parts has specific contribution to the anxiety phenotype.

#### **Natural variants of TPH2 activity**

*TPH2* polymorphisms in humans have been associated with anxiety traits, yet these results have not been supported by other studies. These discrepancies might be partially attributed to the fact that different SNPs in the *TPH2* gene were analyzed. In the mouse *Tph2* gene, a functional SNP has been identified (C1473G) among different inbred mouse strains, which results in the substitution of Pro<sup>447</sup> (1473C allele) with Arg<sup>447</sup> (1473G allele). In mouse strains homozygous for the 1473G allele (G/G), the enzymatic activity of TPH2 was reduced by 50% and correspondingly 5-HT concentrations were found to be decreased in several brain regions.

We provided a detailed comparative neurochemical, molecular, and behavioral characterization of C57BL/6N mice homozygous for either the *Tph2* 1473G or 1473C allele. We showed that this allelic variant alone leads to a reduced *in vivo* 5-HT synthesis rate. However, the distinct and pharmacologically reversible anxiety phenotype in 1473G/G mice is not the result of reduced 5-HT tissue content or 5-HT neurotransmission but is likely mediated via compensatory homeostatic changes involving a functional desensitization of 5-HT<sub>1A</sub>-autoreceptors.

These findings suggest that functional desensitization of 5-HT<sub>1A</sub> autoreceptors is a promising common denominator for increased anxiety.

#### **Targeting the serotonin raphe neurons with new genetic tools**

The generation of mice with overexpression of Tph2 or SERT proved to be impossible, and was abandoned after the generation of 12 transgenic animals. Two alternative approaches were followed. In the first, we investigated the behavioral and neurochemical consequences of a functional C1473G SNP in the mouse *Tph2* gene affecting serotonin synthesis rate. We generated congenic C57BL/6N mice homozygous for the *Tph2* 1473G allele. The Arg(447) substitution in the TPH2 enzyme resulted in a significant reduction of the brain serotonin (5-HT) *in vivo* synthesis rate. Despite decreased 5-HT synthesis, we could detect neither a reduction of brain region-specific 5-HT concentrations nor changes in baseline and stress-induced 5-HT release. However, a functional desensitization of 5-HT(1A) autoreceptors could be identified. Furthermore, behavioral analysis revealed a distinct anxiety phenotype in homozygous *Tph2* 1473G mice, which could be reversed with chronic escitalopram treatment. Alterations in depressive-like behavior could not be detected under baseline conditions or after chronic mild stress. These findings provide evidence for an involvement of functional *Tph2* polymorphisms in anxiety-related behaviors, which are likely not caused directly by alterations in 5-HT content or release but are rather due to compensatory changes

during development involving functional desensitization of 5-HT(1A). (Berger et al., 2012).

The efforts of Partner 6 was redirected towards the production of other transgenic model such as the production of conditional rat transgenic models (Schonig K, et al., BMC Biol. 2012 Sep 3;10(1):77, Conditional Gene Expression Systems in the Transgenic Rat Brain) and models for selective manipulation in serotonergic neurons. (Weber et al., 2009; Weber et al., 2011).

The new mouse and rat driver lines targeting the serotonergic neurons provide useful molecular tools to interrogate the role of widely expressed genes (like vesicular transporter VMAT2, L-type voltage gated calcium channels Cav1.2 and Cav1.3 and glucocorticoid receptor selectively in serotonergic neurons. These tools are now used in our and other labs for such manipulations.

### **Characterizing the Pet1-independent serotonin pathways**

-Pet1 is a transcription factor that plays a key role in the differentiation of serotonin raphe neurons. It is expressed almost exclusively in the raphe of different species and controls together with other transcription factors the serotonin identity. Partner 1a characterized a residual serotonin innervation in the Pet1-KO mice. This led to the individualization of a Pet1-independent serotonin subsystem which targets preferentially limbic areas of the forebrain implicated in stress control, and is characterized by the formation of synaptic junctions. The selective sparing of serotonin innervation in certain regions of the brain was correlated to the analysis of anxiety behaviors in these mice, showing a reduced anxiety in exploration but enhanced fear learning. This work was published: Kiyasova V, et al. J Neurosci. 2011.

-To analyze the physiological and molecular underpinnings of the heterogeneity of the serotonin raphe neurons we carried out a multiscale analysis of single raphe neurons, and anatomical tracing studies. Partner 1a validated the electrophysiological recording of raphe neurons associated with single cell PCR of candidate genes. They also validated a genetic tracing approach using viral constructs in Sert-Cre mice using AAV-containing conditional fluorescent tags. Partner 1a is currently revisiting the anatomy of the ascending B5-B9 raphe cell groups using this approach. The completion of this task is however very time consuming and it will extend beyond the present project in order to finalize a third publication derived from this task.

-As a follow up of this work Partner 1a collaborated with a zebrafish group (Laure Bally-Cuif and Christina Lillesaar) to identify new ETS-factors involved in serotonin specification. This led to identify a new factor ETV5, which appears to be involved in the specification of hypothalamic serotonin neurons. The results of this study are in press in "Development" (Bosco et al. 2013).

### **Complete depletion of serotonin brain levels at different times during development**

Partner1 generated and characterized the VMAT2<sup>flox</sup>:SertCre mice, that had a complete depletion in serotonin stores, and shown that this can be rescued both during development and in adult life, using an inhibitor of monoamine oxidase, clorgyline.

Using this mouse line they showed the important role of central serotonin production in the control of anxiety like behaviors, as the mice were found to have increased flight responses that could be rescued with administration of clorgyline.

Because Sert and VMAT2 have a broad expression during development we further aimed to produce mice with a more specific targeting of VMAT2 deletion to central raphe neurons, and to delimit the timing of this invalidation. Partners 1a and 6 collaborated to further produce two additional mouse strains; The VMAT2::TpH2-Cre-ERT2T enabling a depletion of 5-HT in adult life.

Moreover Partner 1a generated a VMAT2::Pet1-Cre mice, which had not been initially planned. This produced a life-long serotonin depletion but which is limited to the central raphe neurons but did not concern all the raphe neurons. This mouse was characterized biochemically and morphologically.

The VMAT2 models overall allowed to show that central serotonin is needed for the postnatal growth spurt, but not for embryonic/brain growth. (Narboux-Nême et al. in press).

That the cortical development shows only mild retardation in hyposerotonergic mice and the barrel cortex develops normally (Narboux-Nême et al. in press); 3) Adult serotonin depletion rather than developmental effects are implied in the increased escape-like responses observed in the hyposerotonergic mice. (Narboux-Nême et al 2011) 4) Adult serotonin depletion rather than developmental effects are implied in the increased impulsive-like behaviors responses observed in the hyposerotonergic mice. Interestingly, this seems not to be a general increase in impulsivity, but rather a cue-induced change. Animals with VMAT2 knockout show addictive-like behavior which does not depend on reward. 5) Vesicular storage/release is not essential for serotonin neurotransmission. (Narboux –Nême et al. 2011).

### ***WP-2- 5-HT<sub>1A</sub> receptors in anxiety circuits***

The goal of this WP was to identify the neuronal circuits that mediate the effects of the 5-HT<sub>1A</sub> receptor during the maturation of anxiety networks in hippocampus, amygdala, and raphe nuclei. 5-HT<sub>1A</sub> receptor is a clearly identified developmental modulator of anxiety (Gross et al., 2002). Anatomical, biochemical, and physiological defects downstream of this receptor have been identified in previous work by Partner 3. These findings point to a deficit in circuit maturation in the hippocampus of 5-HT<sub>1A</sub> receptor knockout mice. The consortium members will integrate novel approaches to expand and test this hypothesis, taking advantage of anatomical, morphological, and immunohistochemical expertise in the participating laboratories. This animal model will serve as a well-defined example of developmental programming of anxiety and will lead to the detailed identification of the precise neural circuits involved.

### **Generation of transgenic mice expressing 5-HT1A receptor in selected tissues**

Partner 2 (EMBL) produced transgenic mouse lines allowing tissue-specific rescue of 5-HT1A receptors. The following transgenic lines: Htr1a-DG, Htr1a-CeA, Htr1a-RR, Htr1a-CA3, were produced and studied for their anxiety behavior.

Rescue of Htr1a in principal cortical pyramidal neurons (under control of Emx1::Cre tissue-specific driver) was associated with restored anxiety behavior, demonstrating a crucial function for the receptor in these cells for anxiety.

In addition Partner 2 showed that Htr1a autoreceptor and heteroreceptors populations interact to modulate anxiety. Partner 1b (INSERM-LL) observed that flight, but not freezing, behaviors induced by ultrasound (an ethologically relevant aversive cue) are increased in mutant mice lacking the Htr1a receptor (Htr1a knockout) compared to WT mice. Partner 1b (INSERM-LL) further analyzed the behavior of Htr1a knockout mice in the tail suspension test (TST) and the forced swim test (FST). Htr1a knockout mice showed a marked increase of escape-like behaviors in conjunction with a decrease in tonic immobility was observed. In contrast, in the FST no effect of the 5-HT1A receptor deletion was observed. The phenotype of Htr1a knockout mice in the TST was partially reproduced by blockade with blockade of 5-HT1A receptors in the dorsal PAG area. Preliminary data from Partner 1b indicate that flight reactions induced by chemical stimulation of the dorsal PAG are increased in Htr1a knockout mice. Furthermore, in the aversive ultrasound paradigm, these mutant mice displayed increased flight reactions. These data suggest that Htr1a receptors in areas such as the PAG, is a crucial component of defense reactions in some, but not all of the behavioral tests used to screen antidepressant drugs.

Partners 5 (UPO) and 2 (EMBL) have been working during this period on the study of the memory recall and the persistent erasure of hippocampal memory. Following the suggestions given by the Reviewers of the submitted manuscript, both Partners carried out some new experiments. Partner 5 used selective pharmacological tools for the rapid and transient suppression of dentate gyrus granule cells activity. In addition, Partner 2 carried out *in vivo* electrophysiological recordings of granule cells during trace eye-blink conditioning to examine the contribution of dentate gyrus to hippocampal learning and plasticity. In previous experiments, it was concluded that the suppression of DG activity during learning was associated with rapid and persistent memory loss, and it was accompanied by long-term suppression of both conditioned responses and learning-associated synaptic plasticity. Using pharmacological tools, it was tested a possible role of adenosine A1 receptor activation in this synaptic depotentiation. Moreover, some new experiments were designed to demonstrate the generalization of the results found using the eyeblink conditioning paradigm across hippocampus dependent memory tasks (i.e. contextual fear conditioning).

### **Papers published:**

Gross and Canteras, "The Many Paths to Fear" (2012) *Nat Rev Neurosci*. 13:651-8.  
(2012).Gozzi, Jain, et al., "A Neural Switch for Active and Passive Fear" (2010) *Neuron*, 67:656-66. [Reviewed in: H.-C. Pape. (2010) "Petrified or aroused with fear: the central amygdala takes the lead". *Neuron* 67:527-9].

### **Papers submitted:**

Madroñal et al., "Rapid erasure of hippocampal learning and plasticity following transient blockade of dentate gyrus granule cells".

Audero et al., "Suppression of serotonin neuron firing increases aggression in mice"

Piszczyk et al., "Serotonin 1A heteroreceptors and autoreceptors interact to control anxiety in mice".

Piszczyk et al., "Cortical Htr1a receptors modulate anxiety in mice".

Type-I cells in the central nucleus of the amygdala (CeA) tonically bias passive and active fear responses via tonic modulation of lateral CeA projection neurons to forebrain cholinergic. Suppression of serotonin neuron firing does not alter anxiety behavior, but elicits increased aggression.

Htr1a receptors in forebrain and raphe nucleus interact to modulate anxiety, possibly via common action on serotonin homeostasis.

Dentate gyrus granule cells are not necessary for retrieval of hippocampal memory, but are necessary for its acquisition.

Non-dentate gyrus granule cell inputs (EntCtx-CA1) to hippocampus promote depotentiation of learning associated plasticity and conditioned behavior.

Adenosine A1 receptor activation in CA1 is necessary for this depotentiation.

Importantly, the reported effects can be generalized across other hippocampus-dependent memory tasks (i.e., contextual fear conditioning).

The reported findings open the possibility of the targeted erasure of hippocampal memories.

### **Characterization of the phenotype of tissue-specific 5-HT1A receptor rescue mice**

Partner 2 has completed experiments examining hippocampal plasticity in vivo at SC and PP inputs to CA1 in Htr1a-KO mice. These studies revealed no change in plasticity. These data will be included as control experiments in a recently submitted manuscript.

### **Highlight of clearly significant results:**

Submission of manuscript – Gruart et al., "Rapid erasure of hippocampal learning and plasticity following transient blockade of dentate gyrus granule cells"; publication of paper describing role of serotonin in dendritic growth cone dynamics – Ferreira et al., "Serotonin receptor 1A modulates actin dynamics and restricts dendritic growth in hippocampal neurons" *Eur. J. Neurosci.* 2010; submission of manuscript describing structural and electrophysiological deficits in Htr1a-KO mice – Klemenhausen et al., "Altered CA1 hippocampal dendritic morphology and anxiety-like behavior in serotonin-receptor 1A deficient mice".

### ***WP3- 5-HT modulation of GABA circuits***

This work package was intended to identify changes in GABA receptor responses caused by loss of serotonin function. The pilot experiments conducted showed no major changes in GABA circuits or GABA-B receptors caused by disturbed serotonin signaling. Therefore the efforts were redirected towards related questions that emerged during the course of these studies.



### **GABA synapse maturation in the hippocampus and raphe**

Partner 5 analyzed mini-EPSC and mini-IPSC analysis of CA1 pyramidal neurons in Htr1a<sup>KO</sup> mice generated by Partner 2, to evaluate their synaptic architecture and innervation.

Partner 5 started recording of the IPSPs in the hippocampus of behaving mice during classical conditioning of the corneal reflex, with recording in the dentate gyrus, CA3 and CA1 evoked by perforant pathway, Schaffer collateral, and/or commissural inputs. The main objective is to analyse the disynaptic inhibitory synaptic field potentials (fIPSP) which are due to GABA-A and GABA-B effects, respectively. The experiments are in progress.

### **Morpho-functional maturation of GABA interneurons in the hippocampus and amygdala**

-Partner 1a conducted morphological analyses of the GABA interneurons in the hippocampus of Vmat2<sup>Sert/cre</sup> and Pet1<sup>-/-</sup> mice that have respectively a 95% or an 80% depletion of central serotonin levels. Morphological analyses of the hippocampus showed no structural abnormality of the hippocampus. Markers of the GABA interneurons, such as GABA, calbindin and parvalbumin showed comparable distribution in control and KO mice, suggesting that there are no obvious structural alterations in the development of GABA interneurons.

-Partner 1a examined adult neurogenesis in the hippocampus in Pet1 and Vmat2<sup>Sert/cre</sup> mice. This showed significantly increased survival of the newborn dentate granule cells, of both hyposerotonergic mouse strains. This an unexpected finding (given the known enhancement of neurogenesis caused by antidepressants) lead to further analyses to determine the sensitive period for this effect we used the serotonin depleting agent PCPA. This showed that chronic reduction of brain levels of 5-HT in adult- reproduces the phenotype, indicating that this is not a developmental effect

-Partner 5 designed more accurately the experiments proposed in this task, taking in account the results obtained from Partner 1.

### **Identification of KCTD proteins as auxiliary GABA-B receptor subunits**

As an approved deviation (see 2<sup>nd</sup> periodic report Partner 3 investigated the composition of native GABA-B receptor complexes by an unbiased proteomics approach using affinity purification and mass-spectrometry analysis. It was found that GABA-B receptors in the brain are composed of principal and auxiliary subunits (Schwenk et al., 2010, Nature 465, 231-235). The four cytosolic proteins, KCTD8, KCTD12, KCTD12b and KCTD16 tightly associate with the GABA-B core receptors at the plasma membrane and influence agonist potency and the fast kinetics of the receptor response in a KCTD subtype-specific manner. None of the KCTD proteins had been implicated in GPCR function before they were found to be part of GABA-B receptor complexes.

KCTDs are characterized by a common structural motif, the T1 tetramerization domain. In voltage-gated K<sup>+</sup> channels, T1 domains are responsible for the assembly of four subunits around a central channel pore. Likewise, in KCTDs, the T1 domains of four subunits assemble into a homotetramer that tightly binds to the C- terminal domain of GABA-B2. Tyrosine 902 (Y902) in the C-terminal domain of GABA-B2 is required for binding to the T1 domains. Of note, phosphorylation of Y902 is not necessary for KCTD binding, as mutation of Y902 to phenylalanine has no effect on this process. In addition to the T1 domains, KCTD8, KCTD12, KCTD12b and KCTD16 have sequence-related H1 homology domains, with KCTD8 and KCTD16 also featuring sequence-related H2



homology domains. The H1 and H2 domains are not related to each other and exhibit no obvious sequence similarities to other proteins.

When expressed along with GABA-B1 and GABA-B2 in heterologous CHO cells or *X. laevis* oocytes, KCTDs exert a variety of effects on GABA-B receptor-mediated responses. All KCTDs markedly shorten the rise-time of the GIRK-mediated K<sup>+</sup>-current response by up to tenfold, which closely matches the rise-times measured in cultured hippocampal neurons. Moreover, KCTDs differentially increase the potency of GABA at the receptor. Consequently, KCTDs seem to be the missing components that confer fast activation kinetics and distinct agonist potencies to native GABA-B receptors. Additionally, KCTDs may offer an explanation for the variation in the desensitization kinetics of native GABA-B receptor-mediated responses. In the presence of KCTD12, GABA-B receptor activation elicits a strongly desensitizing K<sup>+</sup> current that is characterized by two time constants of 1.5 and 8.9 seconds. Strongly desensitizing K<sup>+</sup> currents are also observed in the presence of KCTD12b. By contrast, in the presence of KCTD8 or KCTD16, the activated receptors induce largely non-desensitizing K<sup>+</sup> currents. Partner 3 recently identified that the KCTD subunit domains exert opposite effects on GABA-B receptor mediated desensitization (Seddik et al., 2012, *J. Biol. Chem.* 287, 39869-39877).

The presence of KCTD12 or KCTD12b also leads to a rapid desensitization of the GABA-B receptor mediated inhibition of voltage-gated Ca<sup>2+</sup> channel (VGCC) currents. Of note, principal and auxiliary GABA-B receptor subunits can be affinity-purified together with native VGCC complexes and, vice versa, These findings show that GABA-B receptors and VGCCs form biochemically stable signaling complexes *in vivo*, and are in line with fluorescence resonance energy transfer (FRET) spectroscopy data demonstrating that GABA-B receptors, G proteins and VGCCs form spatially restricted complexes in the boutons of hippocampal neurons (Laviv et al., 2011, *J. Neurosci.* 31, 12523-12532).

In addition, Partner 3 investigated the temporal and spatial expression patterns of the KCTD proteins in the brain (Metz et al., 2011, *J. Comp. Neurol.* 519, 1435-1454). The results support that most brain GABA-B receptors associate with KCTD proteins, but that the repertoire and abundance of KCTDs varies during development, among brain areas, neuronal populations, and at subcellular sites. This suggests that the distinct spatial and temporal KCTD distribution patterns underlie functional differences in native GABA-B responses.

These exciting findings opened up a new line of research which is partly summarized in a recent review (Gassmann and Bettler, 2012, *Nat. Rev. Neurosci.* 13, 380-394). In summary, the recognition that GABA-B receptors are assembled from principal and auxiliary subunits provided a shift in the understanding of these receptors. It is now clear that molecularly distinct GABA-B receptor subtypes exist, which are distinguished by their auxiliary KCTD subunits. The functional effects of the KCTD proteins together with their distinct spatial and temporal expression patterns, indicates that these auxiliary subunits contribute to the variation in native GABA-B receptor mediated responses during development and in different neuronal populations. Finally, the discovery that KCTDs are auxiliary subunits of GABA-B receptors provides new links between these receptors and disease. In particular, KCTD12 was shown to be a molecular signature of depressive disorders.

### **Modulation of GABA-B signaling by 5-HT**

Partner 3 studied the effects of serotonergic signaling pathways on GABA-B mediated current responses in neurons and transfected CHO cells. GABA-B receptors can be efficiently internalized from the cell surface of cultured neurons by increasing intracellular calcium through activation of either 5-HT<sub>3</sub> or NMDA receptors. However, Partner 3 was unable to demonstrate internalization through 5-HT<sub>3</sub> receptors at synaptic sites and therefore did not pursue this further. In contrast, the experiments with NMDA receptors revealed a clear internalization from dendritic spines, which Partner 3 worked out in detail and recently published (Guetg et al., 2010, Proc. Natl. Acad. Sci. USA 107, 13924-13929). In experiments that were intended to be control experiments for the above experiments Partner 3 identified by serendipity a cross-talk in the GIRK current responses of TAAR1 and dopamine D<sub>2</sub> G-protein coupled receptors. This finding was included in a recent publication (Bradaia et al., 2009, Proc. Natl. Acad. Sci. USA 106, 20081-20086).

### ***WP-4- GABA-B receptors and 5-HT system***

There is strong evidence that a constitutive genetic loss of GABA-B receptors produces an anxiogenic phenotype. GABA-B receptor-deficient mice are more anxious than their wild-type counterparts and show a panic-like response in the elevated zero maze, a variant of the elevated plus maze test (Mombereau et al., 2004, Eur. J. Pharmacol. 497, 119-120). Partner 3 had generated a Cre-conditional allele of the GABA-B<sub>1</sub> gene that allowed site- and time-specific deletion of GABA-B function (Haller et al., Genesis, 40, 125-130). Given that there is a functional interaction between GABA-B and 5-HT<sub>1A</sub> receptors (Luscher et al 1997, Neuron, 19, 687-695), and 5-HT<sub>1A</sub> receptors are also able to regulate anxiety, the hypothesis tested here was that the anxiety phenotype observed in GABA-B-deficient mice was mediated by similar pathways, including the common modulation of brainstem serotonergic neurons.

### **Production of mice with conditional deletion of GABA-B receptors in the raphe**

Partner 3 successfully generated mice that allow for a conditional deletion of GABA-B receptors in serotonergic neurons. These mice are homozygous for the floxed GABA-B<sub>1</sub> allele (provided by Partner 3) and heterozygous for Tph2-CreERT2 (provided by Partner 6). Recombination and successful inactivation of the GABA-B<sub>1</sub> gene following Tamoxifen injection was verified by Partner 3 in electrophysiological recordings (absence of baclofen-mediated K<sup>+</sup>-currents) from serotonergic neurons of the raphe nucleus.

Mice with a conditional deletion of GABA-B receptors in the raphe nucleus were generated and made available to the consortium by Partner 3.

### **Characterization of anxiety behaviors in mice with raphe specific deletion of GABA-B receptors**

Analysis of mice with a conditional deletion of GABA-B receptors in the raphe nucleus did not reveal any significant changes in 5-HT metabolism in serotonergic neurons (Partner 1a and 1b; see below). These findings made it less likely that the conditional KO mice will exhibit an anxiety phenotype. Therefore we concentrated our efforts on the behavioral characterization of mice that allow assessing the contribution of specific GABA-B receptor subtypes and functional states of GABA-B receptors that are controlled by phosphorylation. The results of these studies are described in WP-6.

### **Characterization of the serotonin phenotype in mice with raphe specific deletion of GABA-B receptors**

Partner 1a and 1b analyzed the 5-HT turnover in the raphe-specific GABA-B1 knockout mice by measuring 5-HT and its metabolite 5-HIAA in various brain areas (accumbens, frontal cortex, hippocampus, ventral tegmental area/substantia nigra) using HPLC. The 5-HIAA/5-HT ratio did not show any significant difference between the experimental and control groups of mice. This indicates that GABA-B receptors expressed in serotonergic neurons of the raphe nucleus do not directly contribute to the modulation of 5-HT metabolism in serotonergic neurons. However, a significant decrease in 5-HT turnover was observed in mice lacking the GABA-B1a subunit isoform, but not in mice lacking the GABA-B1b subunit isoform that are predominantly expressed at glutamatergic terminals. Therefore these results identify a GABA-B receptor-mediated modulation of feed forward inhibition of 5-HT neurons in the raphe nucleus. This most likely involves a GABA-B-mediated control of glutamatergic afferents to local GABAergic interneurons regulating the activity of serotonergic neurons. Magnetic resonance spectroscopy, which allows non-invasive measurements of glutamate and glutamine, suggested indeed an increase glutamate/glutamine ratio in the raphe, but not changes in the hippocampus. Microdialysis measurement of extracellular glutamate in living mice is ongoing to corroborate these findings.

At the behavioral level, these changes in glutamate output in the raphe and its consequences on 5-HT turnover at projection sites in the hippocampus are associated with an increase in flight reactions in the aversive ultrasound paradigm, but no increase of anxiety in the social interaction test. These data, presented at the FENS meeting in 2012 in Barcelona, are the subject of publication in preparation.

Partner 1B are also testing whether GABA-A receptors are involved in 5-HT<sub>2C</sub> regulation using both genetic and pharmacological approaches. Pharmacological data indicate that the inhibitory effect of 5-HT<sub>2C</sub> receptors on the stress-induced increase in 5-HT turnover is prevented by the GABA-A receptor antagonist bicuculine. Experiments are on-going to explore whether a similar effect could be mimicked by the constitutive deletion of the  $\alpha 3$  subunit of GABA-A receptors, which is present mainly in monoaminergic neurons. „

### ***WP -5- GABA-B receptors and development***

GABA-B receptors are implicated in the development of anxiety and the control of anxiety states. However, little is known about the developmental time-window, the neuronal systems and the subtypes of GABA-B receptors that are involved in the development of anxiety. Constitutive loss of GABA-B receptors results in increased anxiety, however, acute pharmacological antagonism of GABA-B receptors in adulthood is not anxiogenic (Mombereau et al., 2004). This behavioral profile is similar to that observed with 5-HT<sub>1A</sub> receptor knockout mice (Gross et al., 2002). Thus we wanted to investigate the developmental origins of GABA-B receptor-mediated anxiety and address whether these coincide with alterations in 5-HT function.

### **Characterization of anxiety phenotypes in mice with transient impairment of GABA-B receptor function**

Mice were given GABA-B receptor antagonists and agonist during early life and analysis of behavior will proceed in adulthood. Behavioral experiments completed. Results presented in poster format at EBPS biennial meeting in Rome, September 2009 (Sweeney FF, et al. 2009). This work was also presented as a poster at the 2009 Neuroscience Ireland conference, Trinity College Dublin. A manuscript is currently being prepared for submission to Psychopharmacology.

Partner 5 investigated the consequences of the specific inactivation of the GABA-B1a subunit (mice generated by Partner 3) on the synaptic properties of hippocampal circuits. To this aim learning dependent changes in synaptic strength at the CA3-CA1 synapse was determined by the chronic recordings of fEPSPs evoked across the acquisition and performance phase of an instrumental conditioning task in freely moving mice. The results will be published in a collaborative paper in due course.

### **Tissue-specific and time-specific rescue of GABA-B receptor function**

The individual mouse lines to generate a mouse model allowing tissue- and time-specific rescue of GABA-B function were generated and validated. However, a functional rescue of GABA-B receptors in GABA-B knockout mice was not achieved even though the mice carried all the necessary transgenes. Hippocampal pyramidal neurons of these mice expressed the reporter transgene but not the GABA-B2 subunit necessary for the rescue of GABA-B receptor function. Accordingly, electrophysiological recordings in pyramidal neurons failed to detect functional GABA-B receptors. Therefore a behavioral reversal of the anxiety phenotype in mice with reconstituted GABA-B receptor signaling could not be analyzed.

### **Age-dependent GABA-B-binding characteristics**

Because KCTD proteins increase agonist potency at the receptors we investigated whether these proteins possibly directly increase agonist affinity at the receptor. Using lentiviral vectors we produced stable CHO cell lines co-expressing the core GABA-B receptor subunits GABA-B1 and GABA-B2 together with KCTD8 or KCTD12 to perform <sup>3</sup>H-CGP54626A radioligand antagonist displacement experiments with GABA. At the same time these cell lines were used in GTPγS binding experiments. In addition, radioligand antagonist displacement and GTPγS binding studies in transiently transfected HEK293 cells were performed. The results of these experiments will be published in due course.

## **WP -6- New strategies for treating anxiety**

Partner 3 has generated the pharmacological and genetic tools to validate therapeutic concepts based on GABA-B drugs. While working in the pharmaceutical industry (Novartis, Basel), Partners 3 and partner 4 were involved in the identification and validation of the first generation of positive allosteric modulators at GABA-B receptors (Urwyler et al., 2001; Urwyler et al., 2003; Cryan et al., 2004).

Positive allosteric modulators possess the advantage that they discriminate between activated and non-activated receptor states, while agonists indiscriminately activate all receptors, and may therefore have a broader therapeutic window. Indeed, allosteric modulators of GABA-B receptors confer anxiolytic properties of agonists in the absence of typical side-effects of either agonists or conventional anxiolytics (Cryan et al., 2004).



However, the neural circuits underlying the anxiolytic effects of GABA-B receptor modulators remain elusive. The project of this work package was designed to identify the GABA-B receptor subtypes involved in the activation of anxiety related circuits. Despite some delay in the task due to mouse breeding and import difficulties, significant progress was made. Results were presented at meetings, and several papers are in preparation.

### **GABA-B receptor positive modulator-induced neuronal activation**

Partner 4 (UCC): Mice were given GABA-B receptor positive modulator and exposed to open-arm stress. Brains were processed for c-Fos activation pattern. c-Fos activation pattern was assessed in a broad panel of anxiety-related brain regions. Confirmation of the role of these brain regions in the anxiolytic effects of GS39783 is currently being investigated by directly c-Fos immunohistochemistry was used to map the sites of action of the GABA-B receptor positive modulator GS39783 in the brain in control and stressed animals. Given the new and exciting data on the role of GABA-B receptor subunits in stress-mediation (Task 6.02), we decided to use this same technology to assess the impact whether GABA-B receptor subunits play a role in stress-induced neuronal activation data.

The effects of GABA-B receptor positive modulator on neuronal circuits have now been fully described.

As no data are available in relation to stress reactivity in maternally and non-separated GABA-B1a<sup>-/-</sup> and GABA-B1b<sup>-/-</sup> mice, we analyzed restraint-induced activation pattern of c-fos in various stress-related brain areas.

Given that the hippocampus is involved in the effects of GABA-B receptor ligands and that adult hippocampal neurogenesis is involved in the pathogenesis of stress-related psychiatric disorders we assessed the effects of acute, subchronic and chronic treatment with the GABA-B receptor antagonist CGP52432.

The effects of GS39783 on neuronal activation have been written up for publication and are under revision in Psychopharmacology- Pizzo, RC, O' Leary OF & Cryan JF *Elucidation of the Neural Circuits Activated by a GABA-B Receptor Positive Modulator: Relevance to Anxiety*.

A Review article has been published on the general topic- Cryan JF, Sweeney FF. *The age of anxiety: role of animal models of anxiolytic action in drug discovery*. Br J Pharmacol. 2011 Oct;164(4):1129-61.

Regarding the relative contribution of GABA-B receptors to stress-induced neuronal activation. Our data clearly show that there is a distinct pattern of brain activation between GABA-B1a<sup>-/-</sup>, GABA-B1b<sup>-/-</sup> and wild type mice. Intriguingly, GABA-B1a<sup>-/-</sup> and GABA-B1b<sup>-/-</sup> mice displayed a similar stress-induced expression of c-fos in the cortical areas, in the paraventricular nucleus of the hypothalamus and in the amygdala but not in the hippocampal formation and in the nucleus accumbens. Specifically, GABA-B1b<sup>-/-</sup> mice displayed an increase in stress-induced c-fos expression in the hippocampus and in the nucleus accumbens, two key areas that play an important role in the regulation of antidepressant-like behaviors. Moreover, early-life stress significantly affected stress-

induced c-fos expression in the hippocampus in wild type and GABA-B1b.<sup>-/-</sup> mice but not in GABA-B1a.<sup>-/-</sup> mice.

These data are in preparation for publication- *Felice D, O'Leary OF, Bettler B, Cryan JF. GABA-B receptor subunits modulate stress-induced neuronal activation in the mouse: Impact of Early-Life Stress.*

Chronic, but not acute or subchronic treatment with CGP52432 induced increased cell proliferation in the adult hippocampus. Moreover, these effects were localized to the ventral as opposed to the dorsal hippocampus.

These data have been recently published: *Felice D, O'Leary OF, Pizzo RC, Cryan JF, Blockade of the GABA-B receptor increases neurogenesis in the ventral but not dorsal adult hippocampus: Relevance to antidepressant action, Neuropharmacology 63:1380-1388, 2012.*

### **Receptor subtypes involved in the anxiolytic effects of positive allosteric modulators of R GABA-B**

In order to study which GABA-B receptor subtype mediates the anxiolytic effects of the positive allosteric modulator GS39783, Partner 4 used mice that selectively express either the GABA-B1a or the GABA-B1b subunit. As the wildtype mice failed to show anxiolytic effects of GS39783, it was decided to investigate which GABA-B receptor subunit is responsible for the behavioral effects of cocaine. In addition the GABA-B receptor subunits involved in the effects of chronic psychosocial stress were also investigated (see explanation of the deviation part 3.2 of the Periodic Report).

GABA-B1a and GABA-B1b receptor subunit mutant mice displayed markedly different locomotor responses to both acute and repeated cocaine treatment. GABA-B1a knockout mice displayed enhanced locomotor activity relative to both wildtype and GABA-B1b knockout mice in response to acute cocaine administration. In contrast to this GABA-B1b knockout mice which failed to sensitize to the locomotor effects of cocaine, in contrast to wild type mice which display increasing levels of hyperlocomotor behavior in response to successive doses of cocaine.

Strain and protocol effects were identified that confer increased susceptibility to chronic psychosocial stress were and have been published.

Social defeat stress (SDS) reduced social interaction in wild type and GABA-B1a knockout mice but was without effect in GABA-B1b knockout mice, thus suggesting that GABA-B1b knockout mice are more resilient to the negative effects of chronic social stress on social behavior. SDS reduced the preference for saccharin in GABA-B1a but not in wild type or GABA-B1b knockout mice, thus suggesting that GABA-B1a knockout mice have increased susceptibility to the anhedonic and social effects of social defeat stress.

Taken together, the data suggest that an increased ratio of GABA-B1b receptors to GABA-B1a receptors increases susceptibility to stress-induced depression-like behaviors, while reduced GABA-B1b receptor subunit expression promotes resilience to stress-induced depression-like behaviors.



### **Publications in preparation:**

*O'Leary OF, Felice D, Savignac S, Bettler GABA-B receptor subunits differentially control the susceptibility and resilience to stress-induced depression-related behaviors.*

*Savignac HM, Hyland NP, Dinan TG, Cryan JF. The effects of repeated social interaction stress on behavioral and physiological parameters in a stress-sensitive mouse strain. Behav Brain Res. 2011 Jan 20;216(2):576-84.*

*Savignac HM, Finger BC, Pizzo RC, O'Leary OF, Dinan TG, Cryan JF. Increased sensitivity to the effects of chronic social defeat stress in an innately anxious mouse strain. Neuroscience. 2011 Sep 29;192:524-36.*

*Sweeney F, Jacobson L., Bettler B & Cryan JF Differential role of GABA-B receptor subunits in the behavioral effects of cocaine.*

### **Behavioral characterisation of S892A mice**

Phosphorylation at the S892 residue of the GABA-B2 subunit has been shown in-vitro to modulate receptor desensitization. The effects of S892A mutants in mice (generated by P3) were assessed to determine the functional consequences of this phosphorylation in vivo. In particular animal models of anxiety and cocaine dependence were investigated.

GABA-B2-S892A mice display identical anxiety behavior to wild type littermate controls in the light-dark box, defensive marble burying and stress induced hyperthermia paradigms. In addition they displayed identical behavior to their wild type controls in a Pavlovian fear conditioning experiment with regard to the acquisition, expression and extinction of freezing behavior to a footshock associated cue. The behavior of the mice in the forced swim test, a widely used screen of antidepressant activity, was also identical to wild type controls.

GABA-B-S892A mice displayed identical preference behavior to cocaine as wild type controls in a conditioned place paradigm. Analysis of locomotor activity of mice during this experiment revealed no differences in response to cocaine between the GABA-B-S892A and wild-type mice.

A publication is in preparation - *Sweeney F, Gassmann M, Bettler B & Cryan JF. The phosphorylation site of the S892 residue of the GABA-B2 subunit does not alter behavioral sensitivity to cocaine.*

## **WP -7- Gene-environment effects of 5-HT and GABA-B**

Adverse early life experiences are known risk factors for anxiety in human epidemiological studies; however, the molecular mechanisms underlying these environmental programming effects remain unknown. Several recent studies have identified specific human polymorphisms that are associated with risk for mental illness in the presence of environmental pathogens (so called GxE effects; reviewed in Caspi and Moffitt, 2005). These findings suggest that investigations of genetic susceptibility for anxiety must also consider early environmental risk factors in order to be successful and relevant to a better understanding of human mental illness.

Two paradigms for the manipulation of early environment have been developed by Partner 1b and Partner 2 and were used for testing GxE effects. The first paradigm involved the breeding of mice that have experienced either low or high levels of maternal care (Carola et al., 2006). In humans, poor maternal care is a known risk factor

for anxiety (Pruessner et al., 2005). The second paradigm used the prenatal stress as a model for adverse maternal stress during pregnancy (Vallee et al., 1997).

### **Interactions between BDNF and maternal care on anxiety**

Partner 2 completed experiments documenting the role of Bdnf in the maternal programming of anxiety in (Carola et al., *GBB* 2010) and as well as experiments described in the previous report in mice at postnatal day 10 (Carola et al.). This Partner has also tested for interactions between 5-HTT genotype and chronic psychosocial stress during adulthood (Bartolomucci et al., *Dis. Models and Mech.* 2010).

### **Critical period for the role of 5-HT in moderating the effects of stress on anxiety**

The original plan was to overexpress the Tph2 enzyme which is the rate-limiting enzyme in serotonin synthesis. Partner 6 generated the Tph2-tTA mouse driver line and obtained the tetO-Tph2 founder animals. In parallel, they carried studies using the Tph2-tTa and a reporter gene tetO-lacZ to optimize the induction course of the transgene in serotonergic neurons (Weber et al. 2009). However the doxycycline-regulated gene overexpression did not provide sufficient temporal resolution to determine the critical window so the original aim was abandoned and work efforts were redirected to analyzing the effects of stress in the C1473G SNP. This redirection involved increased efforts from the Partners to use other mouse strains in which there is constitutive depletion of serotonin, but where rescue of the phenotype can be obtained by pharmacological treatments.

-In the course of the work Partners 6 and 1b learned from analysing the C1473G SNP in Tph2, that there are strong regulatory mechanism compensating for variations in Tph2 activity in serotonergic neurons and that such changes play major role in the development and /early postnatal period.

-Partner 1 also characterized the stress responses in mice with constitutive depletion of serotonin but where pharmacological rescue can be obtained; In mice with a deletion of VMAT2 under the control of the SERT promoter. A pharmacological rescue was obtained using clorgyline and in the Pet1-KO mice a rescue of brain serotonin levels was obtained using 5-OH Tryptophan.

### **Assessment of the moderating effects of R GABA-B on early environmental programming of anxiety**

Stress, particularly that in early life is a major predisposing factor for the development of anxiety disorders. We evaluated the contribution of GABA-B1a and GABA-B1b receptor subunit to the effects of low maternal care on behavior.

Efforts were spent on Identifying protocols that generate robust early-life stress induced changes in the mouse. Once a protocol was identified, GABA-B1a<sup>+/+</sup> and GABA-B1b<sup>+/+</sup> mice were bred to generate WT, GABA-B1a<sup>-/-</sup> and GABA-B1b<sup>-/-</sup> mice and genotype was confirmed by PCR. Male and female GABA-B1a<sup>-/-</sup> and GABA-B1b<sup>-/-</sup> mice underwent unpredictable maternal separation combined with unpredictable maternal stress (MSUS) from postnatal day (PND) 1 to 14. During these two weeks maternal care behaviors of the mother were also monitored. During maternal separation, the Ultrasonic vocalizations (USVs) emitted from the maternally separated pups were measured on PND1 and PND7. Upon reaching adulthood the behavior of female and

male maternally-separated (MS) and non-separated (NS) animals were tested in a battery of anxiety and depression tests.

In addition given the relationship between stress anxiety and pain visceral pain responses were assessed in GABA-B1b deficient mice.

**Strain and protocol effects were identified that confer increased resilience to early life stress**

The data investigating the effects of early life stress in GABA-B1 subunit deficient mice revealed a clear gene x environment interaction, with adult MS GABA-B1a<sup>-/-</sup> mice but not MS GABA-B1b<sup>-/-</sup> mice exhibiting anhedonic-like behavior in the saccharin preference and female urine sniffing test. GABA-B1b<sup>-/-</sup> mice exhibited an antidepressant-like behavior in basal conditions as assessed by the tail suspension test and forced swim test. Measurements of anxiety revealed that, GABA-B1b<sup>-/-</sup> pups exhibited an increased number of USVs during MS on PND7. However, in adulthood, the stress induced hyperthermia test of anxiety revealed no differences across all experimental groups. Assessment of locomotor activity in the open field revealed that NS GABA-B1b<sup>-/-</sup> mice displayed increased activity that was attenuated by the MS paradigm, while GABA-B1a<sup>-/-</sup> mice displayed no differences in locomotor activity when compared to the WT group.

These data have been presented at the ECNP meeting in Vienna 2012 as well as other local meetings and are being put together for a publication *Felice D, O'Leary OF, Bettler B, Cryan JF GABA-B receptor subunit isoforms differentially mediate susceptibility to early-life stress-induced depression related behavior.*

Early life stress induced an increase in visceral pain responses in adults. However this was not modulated by the absence of GABA-B1b receptor subunits.

This has recently been published - *Moloney RD, O'Leary OF, Felice D, Bettler B, Dinan TG, Cryan JF. Early-life stress induces visceral hypersensitivity in mice. Neurosci Lett. 2012 Mar 23;512:99-102.*

## 1.4 Potential impact and the main dissemination activities and exploitation of results

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Over 16% of individuals will experience an anxiety disorder during their lifetime (Somers et al., 2006). Recent data within the EU details that the prevalence of anxiety disorders is as high as 41.4 million at an annual cost of €41,372 million (Andlin-Sobocki and Wittchen, 2005). Anxiety disorders include generalized anxiety disorder, panic disorder, social phobia, specific phobia, post-traumatic stress disorder, and obsessive-compulsive disorder (DSM-IVR, 1994). Although there are effective medicines for treating some forms of anxiety (e.g. benzodiazepines, selective-5-HT reuptake inhibitors), most anxiety disorders are poorly treated with these agents due to their side effects, dependence liability or slow onset of action.

Our project provided new information to better understand the neurobiological underpinnings of brain dysfunction in anxiety disorders. The research effort of the consortium provided insights on the genetic and environmental determinants of these affective diseases and how these two major risk factors interact to generate the illness.

### **Scientific Impacts**

-A) The project enabled a better understanding of circuits and systems involved in anxiety disorders. We identified the plasticity mechanism in hippocampal neural circuits involved in associative learning conducting to fear. The use of pharmacogenetic silencing tools allowed to transiently invalidate circuits that are involved in anxiety responses (contextual fear conditioning). We characterized molecular pathways such as the 5-HT<sub>1A</sub> and GABA-B receptors that are involved in the functioning of these circuits.

-B) The project allowed to produce and to characterize new animal models. This included genetically modified animals but also new experimental paradigms to evaluate the neural circuits underlying anxiety disorders. We obtained new tools for targeting the 5-HT raphe neurons and revealed a genetic heterogeneity in the raphe serotonin neurons that would enable to target specific raphe subtypes. We obtained new genetic tools to question the role of the GABA-B receptors at different times in development. These are new scientific tools that will be useful to the community overall, contributing in short and long term to a better understanding of anxiety disorders.

C) The project allowed to identify new therapeutic targets and compounds for translational research. We discovered new pathways of GABA-B signaling: the KCTD proteins, as modulators of GABA-B receptors. These appear promising paths to drug development. We also characterized the cellular and developmental impacts of GABA-B signaling for adult anxiety behaviors.

D) We identified new interactions between genes and the environment in the control of adult anxiety behaviors that could have high relevance to the pathophysiology of anxiety disorders.

E) We identified the role of microbiota in the control of anxiety disorder. This is a completely new dimension in the field that might also be amenable to therapeutic intervention.

### **Impacts in the field of treating disorders**

As mentioned above several of the scientific discoveries cited above could have an impact on therapeutic or preventive approaches in the field of disorders.

As therapeutic approaches one can envisage that better understanding the neural circuits involved in anxiety will lead to a better behavioural control of the anxiety disorders, by stimulating brain plasticity mechanisms in specific manners. At the other end of the spectrum better and more selective drug intervention could be aimed through the discovery of the molecular pathways identified. Finally indirect therapeutic intervention could be sought for such as acting on the composition of the microbiota.

**Prevention:** One of the major aims of the proposal was to define a critical period in the early life, during which modification of gene and/or environment have important consequences in anxiety related disorders. Anxiety disorders share an early onset, mostly before age of 16. Large epidemiological studies have identified prospective risk factors in family history, adverse family environment, and personality traits, suggesting the importance of interactions between genes and environment in developing anxiety disorders. Circumstantial evidence points to the role of early stressful experience in causing modified responses to stress in later life and the role of parental care in harmonious brain development. Our research allowed to determine the role of several molecular signaling pathways, namely downstream of serotonin and GABA neurotransmission that could interact with the environment. This obviously calls to question the use of drugs such as antidepressants or GABA modulators at these critical developmental periods.

### **Impacts for productivity and networking of the European Research effort**

The present proposal exemplified well how through pooling expertise and resources, the critical mass necessary for advancing together better on the scientific issues raised in the project. The exchange of novel information, and the integration of methodologies and new tools was real boost to each of the cooperating institutes. Finally, the possibility of working in parallel on a common task allowed us to achieve realistically the objectives within the time frame of the 7th Framework Programme.

Overall more than 70 scientific articles were published by the teams involved in the consortium. These include high impact publications such as Nature, Nature Neuroscience, Neuron, PNAS and Journal of Neuroscience. These publications have been well received and are well cited by the community, and have often lead to press release communications.

The work was also shown at foremost National and International meetings in the field of neuroscience, such as the FENS and the American Society for Neuroscience, to cite only a few.

Overall the consortium participated to more than 90 such international events where parts of the project could be presented.

### **Impacts for training of young scientists**

Overall a large number of young researchers, Master and PhD students and post-doctoral students received excellent training during this period. The different disciplines covered by the associate Partners enabled 18 students to obtain better knowledge of related fields, and to help them put their own research in a larger perspective.

### **Impact in psychiatry from fundamental research to translational research**

Although this project was essentially fundamental research, it gave impetus to several participating teams to foster their interactions with clinical psychiatrists. For Partner 3 is also co-director of the focal area Neurosciences in the translational Department for Clinical and Biological Research. Partner 1a has contributed to organize a translational network, instance the Paris team has engaged into translational networks with clinical psychiatrists in St Anne in a center for Neurobiology and Psychiatry.



## 1.5 Project public website and contact

The website provides the following information:

1. General information on the project
2. The project presentations
3. The official documents of the project when they are public
4. The work done for each activity of the project through descriptions pages
5. The public data and documents issued by the Devanx work

Website address: <http://devanx.vitamib.com/>

Contact: Isabelle VERDIER, INSERM, France, [Isabelle.Verdier@inserm.fr](mailto:Isabelle.Verdier@inserm.fr)

### **List of all beneficiaries with the corresponding contact name and associated coordinates**

Code	Beneficiary name	Short name	Country	Team Leader	Email
CO01	INSTITUT NATIONAL DE LA SANTE ET DE LA RECHERCHE MEDICALE	INSERM	France	Patricia Gaspar Laurence LANFUMEY	<a href="mailto:patricia.gaspar@inserm.fr">patricia.gaspar@inserm.fr</a> <a href="mailto:laurence.lanfumey@upmc.fr">laurence.lanfumey@upmc.fr</a>
P02	EUROPEAN MOLECULAR BIOLOGY LABORATORY	EMBL	Germany	Cornelius GROSS	<a href="mailto:gross@embl.it">gross@embl.it</a>
P03	UNIVERSITAET BASEL	UNIBAS	Switzerland	Bernhardt BETTLER	<a href="mailto:bernhard.bettler@unibas.ch">bernhard.bettler@unibas.ch</a>
P04	UNIVERSITY COLLEGE CORK, NATIONAL UNIVERSITY OF IRELAND, CORK	UCC	Ireland	John CRYAN	<a href="mailto:J.Cryan@ucc.ie">J.Cryan@ucc.ie</a>
P05	UNIVERSIDAD PABLO DE OLAVIDE	UPO	Spain	Agnès GRUART I MASSO	<a href="mailto:agrumas@upo.es">agrumas@upo.es</a>
P06	ZENTRALINSTITUT FUER SEELISCHE GESUNDHEIT	CIMH	Germany	Dusan BARTSCH	<a href="mailto:Dusan.Bartsch@zi-mannheim.de">Dusan.Bartsch@zi-mannheim.de</a>



## 2. Use and dissemination of foreground

### SECTION A (PUBLIC)

#### A). PUBLIC FINALISED PROJECT OUTPUTS

##### A1). Scientific Publications

LIST OF SCIENTIFIC (PEER REVIEWED) PUBLICATIONS, STARTING WITH THE MOST IMPORTANT ONES											
No.	DOI	Title	Main author	Title of the periodical or the series	Number, date or frequency	Publisher	Place of publication	Date of publication	Relevant pages	Permanent identifiers (if applicable)	Is open access provided to this publication
1		Identifying molecular substrates in a mouse model of the serotonin transporter x environment risk factor for anxiety and depression	C. GROSS	Biol Psychiatry.				01/05/2008			-
2		GABA Homeostasis contributes to the developmental programming of anxiety-related behaviors	C. GROSS	Brain Res	2010	Brain Research Publishing	USA	19/05/2008	189-99	-	No
3		Alpha-Ca <sup>2+</sup> /calmodulin-dependent protein kinase II contributes to the developmental programming of serotonin receptor 1A knock-out mice	C. GROSS	J Neurosci	28(24)	Society for Neuroscience (SFN)	USA	11/06/2008	6250-7	-	No

4		Serotonin transporter transgenic (SERTcre) mouse line reveals developmental targets of serotonin specific reuptake inhibitors (SSRIs)	P. GASPAR	Neuropharmacology	55(6)	-	-	01/11/2008	994-1005		No
5		The sushi domains of secreted GABA(B1) isoforms selectively impair GABA(B) heteroreceptor function	B BETTLER	J Biol Chem	283(45)	American Society of Biochemistry and Molecular Biology Inc.	USA	07/11/2008	31005-11	<a href="http://www.jbc.org/content/283/45/3100">http://www.jbc.org/content/283/45/3100</a>	Yes
6		Inducible gene manipulations in serotonergic neurons	D. BARTSCH	Frontiers in Molecular Neuroscience	02.024.2009	Frontiers Editorial Office	Switzerland	06/11/2009	24	-	No
7		A mouse model for visualization of GABA(B) receptors	JF CRYAN	Genesis	47(9)	-	-	01/09/2009	595-602		No
8		Behavioural and neuroplastic effects of the new-generation antidepressant agomelatine compared to fluoxetine in glucocorticoid receptor-	L. LANFUMEY	Int J Neuropsychopharmacol	13(6)	University of Texas Health Science Centre	USA	24/09/2009	759-74	-	No
9		Conditional gene deletion reveals functional redundancy of GABAB receptors in peripheral nociceptors	B BETTLER	Mol. Pain	5	BioMed Central	USA	19/11/2009	68	<a href="http://www.molecularpain.com/content/5/1/68">http://www.molecularpain.com/content/5/1/68</a>	Yes
10		The selective antagonist EPPTB reveals TAAR1-mediated regulatory mechanisms in dopaminergic neurons	B BETTLER	Proc. Natl. Acad. Sci. USA	106(47)	United States National Academy of Sciences (NAS)	USA	24/11/2009	20081-20086	<a href="http://www.pnas.org/content/106/47/20081.long">http://www.pnas.org/content/106/47/20081.long</a>	Yes

11		The GABAB1a isoform mediates heterosynaptic depression at hippocampal mossy fiber synapses	B BETTLER	J. Neurosci	29(5)	Society for Neuroscience (SFN)	USA	04/02/2009	1414-1423	<a href="http://www.jneurosci.org/cgi/content/full/29/5/1414">http://www.jneurosci.org/cgi/content/full/29/5/1414</a>	No
12		New perspectives on the neurodevelopmental effects of SSRIs	P. GASPAR	Trends in Pharmacological Sciences	31(2)	Cell press	USA	01/02/2010	60-65	-	No
13		Increased vulnerability to psychosocial stress in heterozygous serotonin transporter knockout mice	C. GROSS	J Disease Models Mech	3(7-8)	Company of Biologists Ltd.	USA	01/08/2010	459-70	-	No
14		BDNF moderates early environmental risk factors for anxiety in mouse	C. GROSS	Genes Brain Behav.	9(4)	Wiley-Blackwell and International Behavioural and Neural Genetics Society (IBANGS)	USA	01/01/2010	379-389	-	No
15		Postnatal handling reverses social anxiety in serotonin receptor 1A knockout mice	C. GROSS	Genes Brain Behav.	9(1)	Wiley-Blackwell and International Behavioural and Neural Genetics Society (IBANGS)	USA	01/02/2010	26-32	-	No
16		How can stress alter emotional balance through its interaction with the serotonergic system	L. LANFUMEY	Handbook of Stress: Neuropsychological Effects on the Brain	<a href="http://eu.wiley.com/WileyCDA/WileyTitle/productCd-1444330233.html">http://eu.wiley.com/WileyCDA/WileyTitle/productCd-1444330233.html</a>	Wiley-Blackwell	USA	01/08/2010	-		No
17		The Role of 5-HT2C receptors in the Antidepressant Response: A Critical Review	L. LANFUMEY	Psychopathology and Pharmacotherapy of Depression Mod Trends Pharmacopsychiatry	Mod Trends Pharmacopsychiatry Vol 27	S Karger AG	Switzerland	17/11/2010	155-181		No

18		Native GABAB receptors are heteromultimers with a family of auxiliary subunits	B BETTLER	Nature	465(7295)	Nature Publishing Group	UK	13/05/2010	231-235	<a href="http://www.nature.com/nature/journal/v465/n7295/full/nature08964.html">http://www.nature.com/nature/journal/v465/n7295/full/nature08964.html</a>	No
19		NMDA receptor-dependent GABAB receptor internalization via CaMKII phosphorylation of	B BETTLER	Proc. Natl. Acad. Sci. USA	107(31)	United States National Academy of Sciences (NAS ISA		08/03/2010	13924-9	<a href="http://www.pnas.org/">http://www.pnas.org/</a>	No
		.				)				<a href="http://www.pnas.org/content/107/31/13924.long">content/107/31/13924.long</a>	
20		The sushi domains of GABAB receptors function as dominant axonal targeting signals	B BETTLER	J. Neurosci	30(4)	Society for Neuroscience (SFN)	USA	27/01/2010	1385-1394	<a href="http://www.jneurosci.org/cgi/content/full/30/4/1385">http://www.jneurosci.org/cgi/content/full/30/4/1385</a>	Yes
21		5-HT <sub>2C</sub> receptor activation prevents stress-induced enhancement of 5-HT turnover and extracellular concentration in mice	L. LANFUMEY	J Neurochem	115(2)	-	-	01/10/2010	438-49		No
22		Development of a BAC vector for integration-independent and tight regulation of transgenes in rodents via the Tet system	D. BARTSCH	Transgenic Res	-	Springer	-	18/07/2010	-		No

23	Quantitative analysis of conditional gene inactivation using rationally designed, tetracycline-controlled miRNAs	D. BARTSCH	Nucleic Acids Res	38(17)	Oxford journals	UK	01/09/2010	e168	-	No
24	Genetic models of serotonin (5-HT) depletion: what do they tell us about the developmental role of 5-HT?	S TROWBRIDGE	Anat Rec				03/09/2010			
25	Associative learning and CA3-CA1 synaptic plasticity are impaired in D1R null, Drd1a-/- mice and in hippocampal siRNA silenced Drd1a mice	O.ORTIZ	The Journal of Neuroscience				01/10/2010			
26	The deletion of the microtubule-associated STOP protein affects the serotonergic mouse brain network	V. FOURNET	J NEUROCHEM				01/12/2010			
27	Differential gene expression in mutant mice overexpressing or deficient in the serotonin transporter: a focus on mrocortin 1	V. FABRE	Eur Neuropsychopharmacol.				15/11/2010			
28	Shared changes in gene expression in frontal cortex of four genetically modified mouse models of depression	D. HOYLE	Eur Neuropsychopharmacol.				29/10/2010			
29	Involvement of 5-HT2A receptors in MDMA reinforcement and cue-induced reinstatement of MDMA-seeking behaviour	MJ. OREJARENA	Int J Neuropsychopharmacol.				14/10/2010			

30		GABAB receptors: physiological functions and mechanisms of diversity	A.PINARD	Advances in Pharmacology				2010			
31		Associative learning and CA3-CA1 synaptic plasticity are impaired in D1R null, Drd1a-/- mice and in hippocampal siRNA silenced Drd1a mice	O. ORTIZ	The Journal of Neuroscience				15/09/2010			
32		Motor-coordination-dependent learning, more than others, is impaired in transgenic mice expressing pseudorabies virus immediate-early	JC. LOPEZ-RAMOS	PLoS One;				12/08/2010			
33		The power of reversibility regulating gene activities via tetracycline-controlled transcription	K. SCHONIG	Methods in enzymology				2010			
34		A neural switch for active and passive fear	A.GOZZI	Neuron.				26/08/2010			
35		Serotonin receptor 1A modulates actin dynamics and restricts dendritic growth in hippocampal neurons	TA FERREIRA	Eur J Neurosci.				01/07/2010			
36		A genetically defined morphologically and functionally unique subset of 5-HT neurons in the mouse raphe nuclei	V. KIYASOVA	Neurosc				23/02/2011			



37	Severe serotonin depletion after conditional deletion of the vesicular monoamine transporter 2 gene in serotonin neurons: neural and behavioral consequences	N. NARBOUX-NEME	Neuropsychopharmacology			03/08/2011			
38	Development of raphe serotonin neurons from specification to guidance	V. KIYASOVA	Eur J Neurosci.			21/11/2011			
39	Altered expression of neuronal tryptophan hydroxylase-2 mRNA in the dorsal and median raphe nuclei of three genetically modified mouse models relevant to depression and anxiety	J CHEM NEUROANAT	J Chem Neuroanat.			01/07/2011			
40	Mice with genetic deletion of the heparin-binding growth factor midkine exhibit early preclinical features of Parkinson's disease	RD. PREDIGER	J Neural Transm.			01/08/2011			
41	Effect of enhanced voluntary physical exercise on brain levels of monoamines in Huntington disease mice	T. RENOIR	PLoS Curr.			04/11/2011			
42	Sexually dimorphic serotonergic dysfunction in a mouse model of Huntington's disease and depression	T. RENOIR	PLoS One.			16/07/2011			

43	Functional Tph2 C1473G Polymorphism Causes an Anxiety Phenotype via Compensatory Changes in the Serotonergic System	SM. BERGER	Neuropsychopharmacology				11/04/2011			
44	The oligomeric state sets GABAB receptor signalling efficacy	L. COMPS-AGRAR	The EMBO Journal;				06/05/2011			
45	Distribution of the Auxiliary GABAB Receptor Subunits KCTD8, 12, 12b and 16 in the Mouse Brain	M. METZ	The Journal of Comparative Neurology				01/06/2011			
46	Physical exercise protects against Alzheimer's disease in 3xTg-AD mice	Y. GARCIA-MESA	Journal of Alzheimer's Disease				2011			
47	Role of reuniens nucleus projections to the medial prefrontal cortex and to the hippocampal pyramidal CA1 area in associative learning	E. LYNDELL	PLoS One				15/08/2011			
48	Neurodegeneration and functional impairments associated with glycogen synthase accumulation in a mouse model of Lafora disease	J. VALLES-ORTEGA	EMBO Molecular Medicine				01/11/2011			
49	Learning capabilities and CA1-prefrontal synaptic plasticity in a mice model of accelerated senescence	JC. LOPEZ-RAMOS	Neurobiology of Aging				10/06/2011			

50		Involvement of cannabinoid CB1 receptor in associative learning and in hippocampal CA3-CA1 synaptic plasticity	N. MADROÑAL	Cerebral Cortex				14/06/2011			
51		Validation of the dimensionality emergence assay for the measurement of innate anxiety in laboratory mice	A.JAIN	Eur Neuropsychopharmacol.				23/07/2011			
52		Effect of the interaction between the serotonin transporter gene and maternal environment on developing mouse brain	CV. PASCUCCI	Behav Brain Res.				02/02/2011			
53		Inducible gene manipulations in brain serotonergic neurons of transgenic rats	T. WEBER	PloS one				2011			
54		Early exposure to ethanol differentially affects ethanol preference at adult age in two inbred mouse strains	J. MOLET	Neuropharmacology				01/08/2012			
55		Insights into the complex influence of 5-HT signaling on thalamocortical axonal system development	ES van KLEEF	Eur J Neurosci.				01/05/2012			
56		Probing the diversity of serotonin neurons	P. GASPAR	Philos Trans R Soc Lond B Biol Sci.				05/09/2012			

		TrkB inhibition as a therapeutic target for CNS-related disorders	F. BOULLE	Prog Neurobiol				01/08/2012			
57		Epigenetic regulation of the BDNF gene: implications for	F. BOULLE	Mol Psychiatry				01/06/2012			
58		Impacts of Brain Serotonin Deficiency following Tph2 Inactivation on Development and Raphe Neuron Serotonergic Specification	L. GUTKNECHT	PLoS One				17/08/2012			
59		Beyond the monoaminergic hypothesis: neuroplasticity and epigenetic changes in a	R. MASSART	Philos Trans R Soc Lond B Biol Sci.				05/09/2012			
60		Early exposure to ethanol differentially affects ethanol preference at adult age in two inbred mouse strains	J. MOLET	Neuropharmacology				01/08/2012			
61		Drug withdrawal-induced depression: serotonergic and plasticity changes in animal model	T. RENOIR	Neurosci Biobehav Rev.				01/01/2012			
62		Treatment of depressive-like behaviour in Huntington's disease mice by chronic sertraline and	T. RENOIR	Br J Pharmacol.				01/03/2012			

63		Regulation of neuronal GABAB receptor functions by subunit composition	M. GASSMANN	Nature Reviews Neuroscience			01/06/2012			
64		Early-life stress induces visceral hypersensitivity in mice	RD. MOLONEY	Neuroscience Letters			23/03/2012			
65		Observational learning in mice can be prevented by medial prefrontal cortex stimulation and enhanced by nucleus accumbens stimulation	MT. JURADO-PARRAS	Learning and Memory			18/01/2012			
66		Accelerated aging of the GABAergic septohippocampal pathway and decreased hippocampal rhythms in a mouse model of	SE. RUBIO	FASEB Journal			26/07/2012			
67		Transglutaminase-mediated transamidation of serotonin, dopamine and noradrenaline to fibronectin: Evidence for a general mechanism of	R. HUMMERICH	FEBS Lett.			21/09/2012			
68		Conditional Gene Expression Systems in the Transgenic Rat Brain	K. SCHONIG	BMC Biol			03/09/2012			
69		Tetracycline inducible gene manipulation in serotonergic neurons	T. WEBER	PloS one			31/05/2012			
70		The many paths to fear	CT. GROSS	Nat Rev Neurosci.			01/08/2012			



71		Mouse Models of the 5-HTTLPR × Stress Risk Factor for Depression	CT. GROSS	Curr Top Behav Neurosci.				2012			
72		Investigating anxiety and depressive-like phenotypes in genetic mouse models of	SP. FERNANDEZ	Neuropharmacology				Jan 2012			
73		Development and critical period plasticity of the barrel cortex”	RS. ERZURUMLU	Eur J Neurosci.				May 2012			

## A2). Presentations given at conferences

### A2.1). Presentations – Talks-plenary lectures

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***“How Neural activity controls Development”***, Patricia Gaspar, 10.2007.

*IGBMC, University of Strasbourg, France.*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Role of neurotransmission in shaping cortical maps”***, Patricia Gaspar, 01.2008;

*GraduitenKollege, Mainz, Germany.*

Partner responsible: CO01 INSERM

Type of audience: Scientific community, students.

***“Developmental role of Serotonin”***, Patricia Gaspar, 02.2008.

*Sackler Lecture, University of Toronto, Canada.*

Partner responsible: CO01 INSERM

Type of audience: Scientific community, students.

***“Genetic models to study the developmental role of serotonin”***, Patricia Gaspar, 06.2008.

*7th Dutch Endo-Neuro6Psycho Meeting ; Holland.*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Hyposerotoninergia in VMAT2-conditional KO mice”***, Patricia Gaspar, 07.2008.

*7th Dutch Endo-Neuro6Psycho Meeting ; Holland.*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Multiple tricks of serotonin during development”***, Patricia Gaspar, 09.2008.

*Ecole Lémanique ; Les Diablerets, Switzerland ;*

Partner responsible: CO01 INSERM

Type of audience: Scientific community. Students.

***“Multiple tricks of serotonin during development”***, Patricia Gaspar, 03.2009.

*Graduate course series, University of Edinburgh, UK;*

Partner responsible: CO01 INSERM

Type of audience: Scientific community. Students.

***“Development of cortical sensory maps”***, Patricia Gaspar, 03.2009.

*Graduate closing seminars ED3C, Roscoff, France;*

Partner responsible: CO01 INSERM

Type of audience: Scientific community. Students.

***“Multiple tricks of serotonin during development”***, Patricia Gaspar, 05.2009.

*University of Alicante, Spain ;* Partner responsible: CO01 INSERM

Type of audience: Scientific community. Students.

***“Genetic depletion of serotonin consequence on anxiety”***, Patricia Gaspar, 12.2009.

*Colloque Mediterranéen de Neurosciences, Alexandria , Egypt ;*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Multiple facets of serotonin neurons”***, Patricia Gaspar, 12.2010.

*INMED, Marseille;*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“What Can Be Learned From Hyposerotoninergic Mouse Models”***, Patricia Gaspar, 5.

*1.2011. 26th MORTIMER D. SACKLER Winter Conference In Developmental Psychobiology, Herradura, Costa-Rica.*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Genetic models targeting serotonin systems ; consequences of serotonin depletion”***

Patricia Gaspar, 1.2011.

*University of Baltimore, USA.*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Probing the diversity of serotonin raphe neurons”*** Patricia Gaspar, 5.2011.

*The Neurobiology of Depression-Revisiting the serotonin hypothesis; Montreal, Canada ;*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Serotonin neurons: Investigating the singularities of a diffuse neurotransmitter system”*** Patricia Gaspar, 10/ 2011.

*SFN –satellite - 21st Neuropharmacology Conference “Anxiety and Depression Washington”, USA;*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Serotonin neurons: Investigating the singularities of a diffuse neurotransmitter system”*** Patricia Gaspar, 07/ 2012.

*Stanford University, Palo Alto, USA;*

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Neurogenesis : Basic aspects and unsolved questions. Neuroplasticity – from bench to bedside”***, Laurence Lanfumey, *Journée Scientifique du Centre de Psychiatrie et Neurosciences, Sainte Anne Hospital, Paris (France) May 2010*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Mechanisms of neuroregeneration and neurodegeneration of possible relevance in psychiatric and neurological disorders”***, Laurence Lanfumey, March 2010.

*Euron Workshop on “Drug Treatment of Psychiatric and neurological disorders” University of Minho, Braga (Portugal).*

Partner responsible: CO01b INSERM

Type of audience: Scientific community and Doctorate program

***“What we learned and what can be expected from animal models of depressive disorders”***, Laurence Lanfumey.

*3rd Meeting of West European Societies of Biological Psychiatry Berlin 2-4 June 2010*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Beyond the monoamine hypotheses”***, Laurence Lanfumey.

*EPA Neuroimaging section Imaging Brain Plasticity and development Paris, 1-2 April 2010*

Partner responsible: CO01b INSERM

Type of audience: Scientific community

***“Antidepressant-induced functional adaptive changes in 5-HT<sub>2C</sub> receptors: are they underlying their anxiolytic effects?”*** Symposium, mRNA editing, constitutive activity and therapeutic perspectives of 5-HT<sub>2C</sub> receptors, Raymond Mongeau European College of Neuropsychopharmacology (ECNP) 23rd Congress, Amsterdam, Holland (2010)

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“5-HT<sub>2C</sub> receptor activation inhibits stress-induced increase in 5-HT transmission: relevance to the effects of antidepressant drugs”***, Raymond Mongeau, Symposium, The search for fast-acting antidepressants: An update. 18<sup>th</sup> European Congress of Psychiatry, Munich, Germany (2010)

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Sommes-nous produit de notre histoire ou de nos gènes ? ”***, Laurence Lanfumey.

*Matinées de l'Ireb 12 Oct 2010.*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Neuroplasticité et épigenèse chez la souris”***, Emilien Stragier.

*20<sup>ème</sup> Colloque Scientifique de l'IReB. Créteil, Décembre 2010.*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Epigenetic and clock genes' expression perturbations in genetic and environmental models of depressive-like disorders”***, Renaud Massart.

*7<sup>°</sup> World Congress of Stress. Leiden, The Netherlands Sept 2010.*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Behavioural and neurochemical consequences of 5-HT transporter deletion on 5-HT<sub>2C</sub> receptor functions in mice”***, CBP Martin.

*European College of Neuropsychopharmacology (ECNP) 23rd Congress, Amsterdam, Holland (2010).*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"Beyond monoamine hypotheses : The Neurobiology Of Depression - Revisiting the Serotonin Hypothesis"***, Laurence Lanfumey.

*33e Symposium International du GRSNC Université de Montreal (Can) 2-3 mai 2011*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"Environmental and genetic factors in the aetiology and treatment of anxio-depressive and addictive disorders"***, Laurence Lanfumey.

*University Lille1 CNRS UMR8576, Lille 9 juin 2011.*

Partner responsible: CO01b INSERM

Type of audience: Conference open to general audiences.

***"5-HT receptor modulation of anxiety"***, Laurence Lanfumey.

*European College of Neuropsychopharmacology (ECNP) 24rd Congress, Paris Sept 2011.*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"Pharmacological modulations of epigenetic regulation at BDNF/TrkB signaling in SH-SY5Y cells"***, Fabien Boulle.

*The 9<sup>th</sup> Dutch Endo-Neuro-Psycho (ENP) meeting – Lunteren (Pays-Bas), 1-2 Juin 2011.*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"Relevance of periaqueductal gray 5-HT<sub>1A</sub> receptors in the escape-like behaviors observed in the tail suspension test."***, Raymond Mongeau.

*10th World Congress of Biological Psychiatry. Prague, May 29- June 2 2011*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"Utilisation des tests d'interactions sociales chez la souris dans l'étude des mécanismes d'action des anxiolytiques et des antidépresseurs"***, Raymond Mongeau.

*3ième Congrès Français de Psychiatrie, Symposium, Exclusion sociale et troubles de l'humeur, Lille, France (2011)*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"Pertinence des récepteurs 5-HT<sub>1A</sub> de la substance grise périaqueducule dans les réponses de fuites observées dans le test de suspension par la queue"***, Raymond Mongeau.

*Journées Annuelles de l'AFPN, Paris, 1-2 Avril 2011*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.



***“Contrôle GABAergique du tonus sérotoninergique central-Rôle des récepteurs GABA<sub>B</sub> et implication possible dans l'action antidépressive de la kétamine”,*** CBP Martin.

*Congrès de l'AFPN, Paris, 15-17 mars 2012*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Dépression, au-delà de l'hypothèse monoaminergique”,*** Laurence Lanfumey.

*Université Paris Descartes Centre Universitaire des Saints-Pères Paris 27 mars 2012.*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Alcool : Impact des initiations (trop) précoces : que nous apprend le modèle des rongeurs?”***, Laurence Lanfumey.

*Séminaire du jeudi à la CMME- Service du Professeur Rouillon Paris 02 février 2012.*

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Role of dentate gyrus in hippocampal learning and plasticity”,*** Cornelius GROSS.

*SFN –satellite -Molecular Cellular Cognition (MCCS) Meeting, San Diego, November 12 2010.*

Partner responsible: P02 EMBL

Type of audience: Scientific community.

***“Altered aggressive behavior following genetic and pharmacological manipulation of serotonin autoinhibition”,*** Enrica Audero (winner of the ECNP Fellowship Award for Young Scientist). *European College of Neuropsychopharmacology (ECNP) 23rd Congress, Amsterdam, Holland (2010).*

Partner responsible: P02 EMBL

Type of audience: Scientific community.

***“Altered aggressive behavior following genetic and pharmacological manipulation of serotonin autoinhibition”,*** Enrica Audero (selected speaker), Boris Mlinar, Zhiva K. Skachokova, Antonio Caprioli, Renato Corradetti and Cornelius Gross.

*European college of Neuropsychopharmacology (ECNP) workshop for young scientists in Europe, Nice (France), 4-7 Mars 2010.*

Partner responsible: P02 EMBL

Type of audience: Scientific community.

***“Dissecting Fear and Anxiety”,*** Cornelius Gross

**Reference and place:** Max Planck Institute for Psychiatry, München, April 27, 2010.

Partner responsible: P02 EMBL

Type of audience: Scientific community.

***“Role of hippocampus and amygdala in fear and anxiety”,*** Cornelius Gross

*Italian Institute of Technology, Genova, April 15, 2010.*

Partner responsible: P02 EMBL

Type of audience: Scientific community.

***“Regulation of GABAB receptor signaling by subunit composition”***, Bernhard Bettler, Synaptic Basis of Disease: Translating synaptic physiology into medical applications, University of Geneva, Switzerland, 11-13 July, 2012.

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Regulation of neuronal GABAB receptor functions by subunit composition”***, Bernhard Bettler, Neurobiological Basis of Anxiety Disorders, Institut du Fer à Moulin, Paris, France, June 18, 2012.

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Pharmacology and Physiology of Native GABAB Receptor Heteromultimers”***, Bernhard Bettler, Keystone Symposia on Molecular and Cellular Biology: G Protein-Coupled Receptors: Molecular Mechanisms and Novel Functional Insight, Fairmont, Banff Springs, Alberta, Canada, February 17-22, 2012.

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Regulation of GABAB receptors by auxiliary subunits”***, Bernhard Bettler, Minisymposium Annual Meeting Society for Neuroscience USA 2011, GABA-B receptor signalling in the brain: Insights into plasticity and function, Washington DC, USA, Nov12, 2011

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“GABA<sub>B</sub> receptors as drug targets for mental health disorders”***, Bernhard Bettler, Neurex Workshop “What do genetically modified rodents tell us about neurological and psychiatric disorders?” Institute de génétique et de biologie moléculaire et cellulaire (IGBMC), Illkirch Cedex, France, October 27-28, 2011

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“GABA<sub>B</sub> Receptors - Mechanisms of Diversity”***, Bernhard Bettler, European Symposium on Hormones and Cell Regulation Mont St. Odile, France, October 13-17, 2011

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Beyond GABA<sub>B</sub> receptor heteromers: Insights into the role of auxiliary subunits”***, Bernhard Bettler, 7th International Meeting on Metabotropic Glutamate Receptors, Taormina, Italy, October 2-7, 2011.

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Molecular insights into GABA<sub>B</sub> receptor physiology”***, Bernhard Bettler, European Synapse Summer School 2011, Bordeaux Neuroscience Institute, France, September 4 - 23, 2011.

Title: Molecular insights into GABA<sub>B</sub> receptor physiology

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Molecular insights into GABA<sub>B</sub> receptor functions”***, Bernhard Bettler, *Gordon Research Conference on “Inhibition in the CNS Colby College ,Waterville, Maine, USA, July 24-29, 2011.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Molecular insights into the role of auxiliary GABA<sub>B</sub> receptor subunits”***, Bernhard Bettler, *Symposium “Mechanisms Regulating Membrane Protein Signaling”, The Hebrew University-Hadassah Medical School, Jerusalem, Israel, June 9, 2011.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Patenting inventions arising from biological research: A personal perspective”***, Bernhard Bettler, *NCCR Synapsy – Synaptic Basis of Mental Health Disorders Villars, Switzerland, April 1-2, 2011.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Molecular insights into functionally and pharmacologically distinct GABA-B responses”***, Bernhard Bettler, *Informa Life Sciences Annual G-Protein Coupled Receptors in Drug Discovery Congress Berlin, Germany, March 22-23, 2011.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Reconciling differences between in vitro and in vivo GABA<sub>B</sub> responses”***, Bernhard Bettler, *Danish Society of Neuroscience Spring Symposium, Copenhagen, Denmark, February 4, 2011.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Modulators of GABA-B – Kir3 coupling”***, Bernhard Bettler, *MipTec Conference, Congress Center Basel, Switzerland, September 20-24, 2010.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“Regulation of GABA-B receptor functions by subunit composition”***, Bernhard Bettler, *Paul Scherrer Institute, Villingen, Switzerland, May 23, 2012.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“GABA<sub>B</sub> receptors as drug targets for mental health disorders”***, Bernhard Bettler, *Max-Planck-Institute for Psychiatry, Munich, Germany, October 18, 2011*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“GABA<sub>B</sub> receptors: From structure to synaptic physiology and mental disorders”***

Bernhard Bettler, *University of Regensburg, Germany, July 7, 2011.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***"Molecular insights into GABA<sub>B</sub> receptor physiology"***, Bernhard Bettler, Addex Pharmaceuticals, Geneva, Switzerland, June 16, 2011.

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***"Identification of auxiliary GABA-B receptor subunits"***, Bernhard Bettler, Oregon Health & Science University, Portland, USA, November 18, 2010.

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***"Novel Molecular Mechanisms for Antidepressant Action"***, John Cryan, Karolinska Institute, Sweden, March 2012 Host: Prof Per Svenningsson

Partner responsible: P04 UCC

Type of audience: Scientific community.

***"Stress and the brain-gut axis: From the bowel to behavior"***, John Cryan, 21<sup>st</sup> Neuropharmacology Conference, Washington DC, USA November 2011

Partner responsible: P04 UCC

Type of audience: Scientific community.

***"mGluR7: a therapeutic target at the interface of cognition and emotion"***, John Cryan, 14<sup>th</sup> biennial European Behavioural Pharmacology Society Meeting, Amsterdam August 2011

Partner responsible: P04 UCC

Type of audience: Scientific community.

***"Stress and the Brain-Gut Axis: From Bowel to Behaviour"***, John Cryan, NUI Galway, Galway, Ireland December 2011

Partner responsible: P04 UCC

Type of audience: Scientific community.

***"Exciting times beyond the brain: Peripheral mGluRs"***, John Cryan, 7<sup>th</sup> International Meeting of Metabotropic Glutamate Receptors in Taormina, Sicily, Italy, October 2011.

Partner responsible: P04 UCC

Type of audience: Scientific community.

***"Genetic and environmental studies into the role of GABAB receptors in anxiety"***, John Cryan, Univ Wurzburg, Germany, September 2011

Partner responsible: P04 UCC

Type of audience: Scientific community.

***"Microbiome-Gut-Brain-Axis: From Bowel to Behaviour"***, John Cryan, Korea Yakult Co. Ltd. Yongin, South Korea, August 2011

Partner responsible: P04 UCC

Type of audience: Scientific community.

***"Stress and the Brain-Gut Axis: From the Bowel to Behaviour"***, John Cryan, University

*of Tartu, Estonia, November, 2010,*  
 Partner responsible: P04 UCC  
 Type of audience: Scientific community.

***“Stress and the Brain-Gut axis: Neural mechanisms and genetic influences”***, John Cryan, 7<sup>th</sup> World Congress on Stress, Leiden, The Netherlands, August 2010  
 Partner responsible: P04 UCC  
 Type of audience: Scientific community.

***“Animal models of co-morbid depression and irritable bowel syndrome”***, John Cryan, 23<sup>rd</sup> Congress of European College of Neuropsychopharmacology, Amsterdam, The Netherlands, August 2010.  
 Partner responsible: P04 UCC  
 Type of audience: Scientific community.

***“Bowel to Behaviour: Immune regulation of the brain-gut axis”***, John Cryan, 17<sup>th</sup> Annual Psychoneuroimmunology Research Society Meeting, Dublin, Ireland From,  
 Partner responsible: P04 UCC  
 Type of audience: Scientific community.

***“From Bowel to Behaviour- Stress-based models of co-Morbid Depression and Irritable Bowel Syndrome”***, John Cryan,, University of Regensburg, Germany, February 2010  
 Partner responsible: P04 UCC  
 Type of audience: Scientific community.

***“Silencing the Unquiet Mind: Neuronal siRNA delivery in vivo and its application to Neuropsychiatric disorders”***, John Cryan, University of Leeds, Faculty of Biological Sciences Seminar series, February 2010..  
 Partner responsible: P04 UCC  
 Type of audience: Scientific community.

***“Learning as a functional state of the brain: studies in wild-type and transgenic animals”***, José M. Delgado-García, IGBMC, University of Strasbourg, France, 13.01.2011.  
 Partner responsible: P05 UPO  
 Type of audience: Scientific community.

***“Neurobiología del aprendizaje asociativo”***, Agnès Gruart, Instituto Politécnico Nacional. Superior School of Medicine, México, D.F. (México), 10.03.2011.  
 Partner responsible: P05 UPO  
 Type of audience: Scientific community.

***“Mecanismos funcionales del aprendizaje motor y cognitivo”***, Agnès Gruart, Faculty of Medicine, University Complutense of Madrid (Spain), 28.04.2011.  
 Partner responsible: P05 UPO  
 Type of audience: Scientific community.

***“Physiology and pathology of the eyelid motor system”***, José M. Delgado-García, Università degli Studi di Milano, Italy, 17.06.2011.



Partner responsible: P05 UPO

Type of audience: Scientific community; Doctorate program

***“Circuitos neuronales que generan el aprendizaje asociativo”***, Agnès Gruart, Faculty of Medicine. *University of Castilla La Mancha, Ciudad Real (Spain), 23.10.2011.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Functional states underlying associative learning in mammals”***, José M. Delgado-García, *The Max Planck Institute for Medical Research, Heidelberg, Germany, 13.01.2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Introducción a los mecanismos de aprendizaje y memoria”***, José M. Delgado-García, *Faculty of Medicine, Salamanca University, Spain. 02.02.2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community and Doctorate program

***“Cortical basis of associative learning”***, Agnès Gruart, *Friederich Miescher Institute for Biomedical Research, Basel (Switzerland), 3.05.2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Workshop of the 7th Framework Programme: Health”***, Agnès Gruart, *University of Córdoba and Andalusian Government, Córdoba (Spain), 15.07.2010.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Mecanismos neuronales que subyacen al aprendizaje, la creatividad y el recuerdo”***  
José M. Delgado-García. *Menendez Pelayo International University, Santander, Spain, 07.2012;*

Partner responsible: P05 UPO

Type of audience: Conference open to general audiences.

***“Mechanisms for memorizing and forgetting”***, Agnès Gruart, *National Royal Academy of Medicine, Madrid (Spain), 9.02.2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Gene und Gedächtnis: was können wir von konditionalen Maus-Mutanten lernen?”***

Dusan Bartsch, 2.2008. *University Kaiserslautern, Kaiserslautern, Germany.*

Partner responsible: P06 CIMH

Type of audience: Scientific community.

***“Conditional genetic manipulations in serotonergic neurons”***, Dusan Bartsch, 4.2008. *Columbia University, USA.*

Partner responsible: P06 CIMH

Type of audience: Scientific community.

***“Conditional expression systems in transgenic rodents”***, Dusan Bartsch 6.2009. *University Mainz, Germany.*



Partner responsible: P06 CIMH  
Type of audience: Scientific community.

**“Genetics of Alcohol Addiction”**, Dusan Bartsch, 11.2009. *NGFN Plus, Berlin, Germany.*

Partner responsible: P06 CIMH  
Type of audience: Scientific community.

**“Conditional RNAi-based knockdown”**, Dusan Bartsch , 3.2010. *Abbott, Ludwigshafen, Germany.*

Partner responsible: P06 CIMH  
Type of audience: Scientific community.

**“Age-dependent Memory Impairment”**, Dusan Bartsch, 9.2010. *Heidelberg University, Heidelberg, Germany.*

Partner responsible: P06 CIMH  
Type of audience: Scientific community.

**“Conditional gene control in the rodent brain”**, Dusan Bartsch, 3.2011. *Academy of Sciences, Praha, Czech Republic*

Partner responsible: P06 CIMH  
Type of audience: Scientific community.

**“Transgenesis Strategies”**, Dusan Bartsch, 8.2011. *Academy of Sciences, Praha, Czech Republic*

Partner responsible: P06 CIMH  
Type of audience: Scientific community.

**“RNAi-based knockdown in transgenic rodents”**, Dusan Bartsch, 9.2011. *University Magdeburg, Magdeburg, Germany*

Partner responsible: P06 CIMH  
Type of audience: Scientific community.

**“Forebrain-specific knockdown of Staufen2 in transgenic rats impairs synaptic plasticity and learning and memory processes”**

S. BERGER<sup>1</sup>, I. FERNÁNDEZ-LAMO<sup>2</sup>, K. SCHÖNIG<sup>1</sup>, S. CLEMENTI<sup>1</sup>, M. KIEBLER<sup>3</sup>, A. KONECNA<sup>3</sup>, S. GROTHE<sup>4</sup>, O. VON BOHLEN UND HALBACH<sup>4</sup>, J. M. DELGADO<sup>2</sup>, A. GRUART<sup>2</sup>, D. BARTSCH<sup>1</sup>; . Society for Neuroscience, New Orleans, USA Oct, 2012.

Partner responsible: P06 CIMH  
Type of audience: Scientific community.

## **A2.2). Presentations – Posters**

**“Genetic heterogeneity of serotonin neurons in the raphe revealed in Pet1-KO mice”**

Kiyasova V., Stankovski L., Muzerelle A. , Deneris E. & Gaspar, *Society for Neuroscience-Chicago, USA Oct 21, 2009.*

Partner responsible: P01a INSERM  
Type of audience: Scientific community.

***“Conditional vesicular monoamine transporter 2 knockout: A new tool to study the developmental and behavioral consequences of central serotonin depletion”***

GASPAR, N. NARBOUX-NÊME, R. MONGEAU, G. ANGENARD, C. SAGNÉ. GIROS, M. HAMON, L. LANFUMEY. *Society for Neuroscience-Chicago, USA Oct 21, 2009.*

Partner responsible: P01a INSERM

Type of audience: Scientific community.

***“Genetic heterogeneity of raphe serotonin neurons as revealed in Pet1-KO mice”***

Kiyasova V. , Fernandez S. P. , Laine J., Stankovski L. , Muzerelle A. , Deneris E. & Gaspar P. *7th FENS Forum - Amsterdam, July 2010.*

Partner responsible: P01a INSERM

Type of audience: Scientific community.

***“Serotonin modulation of anxiety, fear and memory as revealed by Pet1-KO mice”***

Fernandez S. P., Kiyasova V & Gaspar P. *7th FENS Forum - Amsterdam, July 2010.*

Partner responsible: P01a INSERM

Type of audience: Scientific community.

***“Altered cortical maturation but normal barrel development in hyposerotonergic mice”***

Gaspar P., Lanfumey, L ; Mongeau, R. Narboux-Nême, N. *7th FENS Forum - Amsterdam, July 2010.*

Partner responsible: P01a INSERM

Type of audience: Scientific community.

***“Paradoxical increase of hippocampal neurogenesis in serotonin deficient mice”***

S. K. Trowbridge, N. Narboux-Neme, S. L. Diaz, S. Jessberger, E. S. Deneris, P. Gaspar; *Society for Neuroscience- San Diego Monday, Nov 15, 2010.*

Partner responsible: P01a INSERM

Type of audience: Scientific community.

***“Identifying subclasses of ascending raphe neurons”***

S Fernandez, C Cabezas, B Cauli, P Gaspar

*Society for Neuroscience- Washington, October, 2011.*

Partner responsible: P01a INSERM

Type of audience: Scientific community.

***“Multiscale analysis of single raphe neurons”***

S Fernandez, C Cabezas, B Cauli, P Gaspar

*IUPHAR, Serotonin Club Meeting, Montpellier, July 2012.*

Partner responsible: P01a INSERM

Type of audience: Scientific community.

***“Mice with decreased vesicular glutamate transporter VGLUT1 levels show an altered presynaptic 5-HT function”***

Garcia-Garcia AL, Elizalde N, Venzala E, Del Rio J, Lanfumey L, Tordera RM. *European Neuropsychopharmacology. 2010 March;20:S9-S10.*

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“Behavioral and neurochemical consequences of 5-HT transporter deletion on 5-HT(2C) receptor functions in mice”***

Martin C, Chevarin C, Lesch KP, Hamon L, Lanfumey L, Mongeau R. *European Neuropsychopharmacology*. 2010 March;20:S44-S.

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“Involvement of 5-HT<sub>2A</sub> receptors in MDMA reinforcement and cue-induced reinstatement of MDMA seeking behaviour”***

Program N° 270.2/JJ8, Neuroscience Meeting Planner. Robledo P, Orejarena MJ, Lanfumey L, Maldonado R, *San Diego CA: Society for Neuroscience, 2010. Online*

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“Anxiety-like and aggressive behaviors in mutant mice with fully edited 5-HT<sub>2C</sub> receptors”***

Martin CBP, Hamon M, Lanfumey L, Mongeau R.

*10<sup>e</sup> Colloque des Neurosciences Marseille, May 24-27, 2011*

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“Epigenetic modifications in genetic and environmental mouse models of depressive-like disorder: reversal by chronic treatment with agomelatine”***

Massart R, Stragier S, Païzanis E, Gabriel C., Mocaer E., Hamon M., Lanfumey L. *Program No. 394.18/WW16 2011 Neuroscience Meeting Planner. Washington, DC: Society for Neuroscience, 2011. Online*

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“Yif1B, a chaperone protein for the dendritic targeting of the serotonin 5-HT<sub>1A</sub> receptor: from brain localization to knock-out mice”***

Masson J, Franck B, Bazin le Douarin N, Lanfumey L, Darmon M. *10<sup>e</sup> Colloque des Neurosciences Marseille, May 24-27, 2011*

Partner responsible: P01bINSERM

Type of audience: Scientific community.

***“Relevance of periaqueductal gray 5-HT<sub>1A</sub> receptors in the escape-like behaviors observed in the tail suspension test”***

Mongeau R, Daran A, Lanfumey L, *10th World Congress of Biological Psychiatry. Prague, May 29- June 2 2011*

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“Epigenetic modifications after voluntary and chronic alcohol intake in C57BL/6J mice”***

Stragier E, Massart R, Hamon M, Lanfumey L.

*“Epigenetics, Brain and Behavior”, Colloques “Médecine et Recherche” Fondation Ipsen Paris - April 18, 2011*

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“GABAergic control of central serotonergic neurotransmission – Role of GABA<sub>B</sub> receptors and possible implication in the antidepressant action of ketamine”***

Martin CBP, Chevarin C, Gassman M, Hamon M, Bettler B, Lanfumey L, Mongeau R.. *The 8<sup>th</sup> FENS Forum of Neuroscience, Barcelona (Spain), 14-18 juillet 2012.*

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“Chronic ethanol intake-induced epigenetic modifications at bdnf gene level in the mouse hippocampus”***

Stragier E, Massart R, Hamon M, Lanfumey L. *The 8<sup>th</sup> FENS Forum of Neuroscience, Barcelona (Spain), 14-18 juillet 2012.*

Partner responsible: P01b INSERM

Type of audience: Scientific community.

***“Functional connectivity in descending hippocampal projections to cortex and septum during anxiety in mice ”***

N. MADRONAL, B. SILVA, C. GROSS. *Society for Neuroscience-Washington, USA Oct , 2011.*

Partner responsible: P02 EMBL

***“A mouse model of the human 5-HTT-LPR polymorphism”***

L. P. PISZCZEK, W. GRAJ, K. SCHLAX, O. ERMAKOVA, C. GROS. *Society for Neuroscience-Washington, USA Oct , 2011.*

Partner responsible: P02 EMBL

***“Selective expression of serotonin 1A autoreceptors in transgenic mice does not influence anxiety”***

C. T. GROSS, L. P. PISZCZEK, K. SCHLAX, E. AUDERO, B. MLINAR, G. BACCINI, R. CORRADETTI. *Society for Neuroscience-Washington, USA Oct , 2011.*

Partner responsible: P02 EMBL

***“Dissecting the role of serotonin 1A hetero- and autoreceptors using a novel Cre-conditional allele ”***

L. P. PISZCZEK, K. SCHLAX, A. WYRZYKOWSKA, A. PISZCZEK, E. AUDERO, C. GROSS. *Society for Neuroscience-New Orleans, USA Oct , 2012.*

Partner responsible: P02 EMBL

***“Changes of local field potential rhythmic activities in hippocampus and prefrontal cortex by benzodiazepine anxiolytics ”***

Zhan, Y, Gross C.. *Society for Neuroscience-New Orleans, USA Oct 15, 2012.*

Partner responsible: P02 EMBL

***“Native GABAB receptors are heteromultimers with a family of auxiliary subunits”***

Martin Gassmann: *40<sup>th</sup> annual Meeting Neuroscience 2010, St. Diego, USA, 2010.*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“GABA-B receptor subtypes as therapeutic targets for mental health disorders”***

R. Turecek, B. Bettler: *NCCR Synapsy Annual Meeting, 30-31.03.2012, Villars, Switzerland*

Partner responsible: P03 UNIBASEL

Type of audience: Scientific community.

***“GABA-B receptor subunit isoforms differentially mediate susceptibility to early-life stress-induced depression related behavior”***

*D. Felice, O. F. O’Leary, B. Bettler, J. F. Cryan (2012), 25<sup>TH</sup> ECNP CONGRESS, VIENNA OCTOBER 2012*

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“GABAB(1) Receptor Isoforms Play a Differential Role in Susceptibility to Anxiety and Depression-Related Behaviours following Early Life Stress”***

*D. Felice, O. F. O’Leary, B. Bettler, J. F. Cryan. Pharmacy All Ireland Schools Conference, March 2012, Cork, Ireland*

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“GABAB(1) receptor subunit isoforms differentially mediate susceptibility to depression-related behaviour following early-life stress”***

*D. Felice, O. F. O’Leary, B. Bettler, J. F. Cryan. ; ECNP Workshop on Neuropsychopharmacology for Young Scientists in Europe, 15-18 March 2012, Nice, France*

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“GABAB(1) receptor isoforms confer differential susceptibility to the effects of social defeat stress Anxiety and Depression”***

*O’Leary OF, Savignac HM, Felice D, Bettler B, Cryan JF (2011), 21st Neuropharmacology Conference, Virginia, USA*

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“GABAB(1) receptor isoforms confer differential susceptibility to the effects of social defeat stress in mice”***

*O’Leary OF, Savignac HM, Bravo JA, Vaugeois JM, Bettler B, Cryan JF (2011). 6th Annual Meeting of Neuroscience Ireland, Dublin, P1.40:*

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“Adult Hippocampal Neurogenesis is Increased by a GABA<sub>B</sub> Receptor Antagonist in Balb/c Mice: Relevance to Antidepressant Action”***

*D. Felice, O. F. O’Leary; R. C. Pizzo; J. F. Cryan (2011).. Poster, Anxiety and Depression, Neuropharmacology Conference, 10-11 November, Washington, USA*

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“The Role of GABA<sub>B(1)</sub> Receptor Subunits in the Modulation of the Effects of Early Life Stress on Anxiety and Depression-Related Behaviour in Adulthood, Poster, Anxiety and Depression”***

*D. Felice, O. F. O’Leary, B. Bettler, J. F. Cryan (2011)., Neuropharmacology Conference, 10-11 November, Washington, USA*

Partner responsible: P04 UCC



Type of audience: Scientific community.

***“Adult Hippocampal Neurogenesis is Increased by a GABA<sub>B</sub> Receptor Antagonist in Balb/c Mice: Relevance to Antidepressant Action”***

D. Felice, O. F. O’Leary; R. C. Pizzo; J. F. Cryan (2011). Poster, Neuroscience Ireland, September 1st-2<sup>nd</sup>, Maynooth, Ireland

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“The Role of GABA<sub>B(1)</sub> Receptor Subunits in the Modulation of the Effects of Early Life Stress on Anxiety and Depression-Related Behaviour in Adult Mice”***

D. Felice, O. F. O’Leary, B. Bettler, J. F. Cryan (2011)., Poster, Neuroscience Ireland, September 1st-2<sup>nd</sup>, Maynooth, Ireland

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“GABA<sub>B</sub> receptor blockade induces antidepressant-like behavioural effects: Relationship with Neurogenesis. European Behavioural”***

D. Felice, O.F. O’Leary, F. Sweeney, R.C.Pizzo and J.F.Cryan (2010). Pharmacology Society, Workshop on drugs, psychiatric disorders and neurogenesis, September 2nd-3rd, Tours, France. Behavioural Pharmacology 21(5-6):589.

Partner responsible: P04 UCC

Type of audience: Scientific community.

***“El ejercicio físico revierte parcialmente los déficits de plasticidad sináptica en el ratón 3XTG-AD”,***

López-Ramos, J.C., García, Y., Guerra, R., Gruart, A., Giménez-Llort, L., Sanfeliu, C. and Delgado-García, J.M., Salamanca (Spain): 4 National Meeting of the Spanish Society of Medicine, 2010.

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Fenotipaje cognitivo y estudio electrofisiológico de ratones SAMP8 como modelo de envejecimiento acelerado”,***

López-Ramos, J.C., Sanfeliu, C., Acuña-Castroviejo, D., Gruart, A. and Delgado-García, J.M., Valladolid (Spain): Meeting of the Spanish Society of Geriatric and Gerontology, 2010.

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Local CB1 hippocampal receptor activation is sufficient for reproducing the effects of cannabinoids on associative learning”,***

Madroñal, N., Gruart, A., Valverde, O., Moratalla, R. and Delgado-García, J.M., Amsterdam (Holland): 7<sup>th</sup> Forum of European Neuroscience (FENS), 2010.

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Changes in strength at the hippocampal CA3-CA1 synapse during operant conditioning in behaving mice”,***



Jurado-Parras, M.T., Vega-Flores, G., Castellanos, N.P., del Pozo, F., Gruart, A. and Delgado-García, J.M., *Amsterdam (Holland): 7<sup>th</sup> Forum of European NEuroscience (FENS), 2010.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

**“Plastic modifications induced by object recognition memory processing”,**

Clarke, J., Cammarota,

M., Gruart, A., Izquierdo, I. and Delgado-García, J.M., *Amsterdam (Holland): 7<sup>th</sup> Forum of European NEuroscience (FENS), 2010.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

**“Early effects of choline (CHOL) and uridine-monophosphate (UMP) on instrumental conditioning and long-term potentiation (LTP) in rat pups”,**

Ramírez, M., Barranco, A., Jiménez, M.L., Martín, M.J., Oliveros, E., Gruart, A., Delgado-García, J.M. and Rueda, R., *Sevilla (Spain): 43<sup>rd</sup> European Brain and Behaviour Society Meeting (EBBS), 2011.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

**“Hippocampal mechanisms of self-stimulation behavior”,**

Vega-Flores, G., Gruart, A., López, I. and Delgado-García, J.M., *Sevilla (Spain): 43<sup>rd</sup> European Brain and Behaviour Society Meeting (EBBS), 2011.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

**“Driving cognitive processes by brain stimulation in mice”,**

Jurado-Parras, M.T., Gruart, A. and Delgado-García, J.M., *Sevilla (Spain): 43<sup>rd</sup> European Brain and Behaviour Society Meeting (EBBS), 2011.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

**“Hippocampal mechanisms of learning and preference during septal brain stimulation reward”,**

Vega-Flores, G., Gruart, A. and Delgado-García, J.M., *Barcelona (Spain): 8<sup>th</sup> FENS Forum of Neuroscience, 2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

**“Forebrain-specific knockdown of Stauf2 in transgenic rats impairs synaptic plasticity, spatial working memory and spatial novelty detection”,**

Clementi, S., Berger, S., Schoening, K., Gruart, A., Delgado-García, J.M. and Bartsch, D., *Barcelona (Spain): 8<sup>th</sup> FENS Forum of Neuroscience, 2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

**“Erasure of hippocampal learning and plasticity following transient blockade of dentate gyrus”,**

Madroñal, N., Gruart, A., Jan, A., Fernández-Guizán, A., Tsetsenis, T. and Gross, C., *Barcelona (Spain): 8th FENS Forum of Neuroscience, 2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Brain Stimulation Reward and Hippocampus”***

Vega-Flores, G., Gruart, A. and Delgado-García, J.M., *Barcelona (Spain): 8th FENS Forum of Neuroscience, 2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Role of prefrontal circuits in decision making tasks involving different environmental constraints”***

Jurado-Parras, M.T., López-Ramos, J.C., Guerra-Narbona, R., Gómez-Climent, M.A., Hernández-González, S., Gruart, A. and Delgado-García, J.M., *Barcelona (Spain): 8th FENS Forum of Neuroscience, 2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community.

***“Multisynaptic state functions characterizing the acquisition of new motor and cognitive skills”***

Delgado-García, J.M., Sánchez-Campusano, R., Carretero-Guillén, A., Fernández-Lamo, I., Muñoz, M.D. and Gruart, A., *Barcelona (Spain): 8th FENS Forum of Neuroscience, 2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Observational learning in mice can be prevented by medial prefrontal cortex stimulation and enhanced by nucleus accumbens stimulation”***

Gruart, A., Jurado-Parras, M.T., Carponcy, J. and Delgado-García, J.M., *Barcelona (Spain): 8th FENS Forum of Neuroscience, 2012.*

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Erasure of hippocampal learning and plasticity following transient blockade of dentate gyrus”***

N. MADRONAL, J. DELGADO-GARCÍA, A. FERNÁNDEZ-GUIZÁN, J. CHATTERJEE, M. KÖHN, A. JAIN<sup>1</sup>, T. TSETSENIS, C. GROSS, A. GRUART. *Society for Neuroscience-New Orleans, USA Oct, 2012.*

Partner responsible: P05 UPO

***“Behavioral analysis of mice with embryonal and adult deletion of the glucocorticoid receptor in serotonergic neurons”***

S. WIRTH<sup>1</sup>, T. WEBER<sup>1</sup>, S. KUTSCHERJAWY<sup>1</sup>, M. VOGT<sup>2</sup>, S. BERGER<sup>3</sup>, G. SCHÜTZ<sup>3</sup>, P. GASS<sup>2</sup>, D. BARTSCH<sup>1</sup>, S. BERGE. *Society for Neuroscience Washington, USA Nov, 2011.*

Partner responsible: P06 CIMH

***Fate Mapping and Manipulation of Adult Neurogenesis***

Dusan Bartsch, 9.2011. *Euro Glia, Praha, Czech Republic*

Partner responsible: P06 CIMH

Type of audience: Scientific community.

**A Functional *Tph2* C1473G Polymorphism Causes an Anxiety Phenotype via Compensatory Changes in the Serotonergic System**

Dusan Bartsch, 5.2012. *NGFN*, FU Berlin, Berlin, Germany

Partner responsible: P06 CIMH

Type of audience: Scientific community.

**A3). Origination of seminars; symposia**

***“Symposium at the 9<sup>th</sup> Colloquium of the French Society for Neuroscience”*** Bordeaux, May 26 – 29, 2009; France 3 Researchers of the DEVANX made presentations at this symposium.

Partner responsible: Partner 01 INSERM organized the symposium

Type of audience: Scientific community

***“Symposium along the Devanx scientific theme for the Third Mediterranean Conference of Neuroscience”***, Egypt, Alexandria, December 2010.

Partner responsible: Partner 01 INSERM co-organized the symposium

Type of audience: Scientific community.

***“The Neurobiological basis of anxiety disorders”***, June 2012, Institut du Fer à Moulin. All the Devanx partners participated, with additional speakers.

Partner responsible: Partner 01 INSERM organized the one day Final meeting on the Devanx project

Type of audience: Scientific community, Students.

***“Differing contributions of hippocampal synapses to associative learning tasks”***, Agnès Gruart, Carmona (Sevilla, Spain): 9<sup>th</sup> Management Committee Meeting and 9<sup>th</sup> Working Groups Meeting. COST B30: Neural Regeneration and Plasticity (NEREPLAS), 2010.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Learning mechanisms studied in wild-type and transgenic mice”***, Agnès Gruart, Toulouse (France): *Un Symposium en Hommage à Jean-Michel Lassalle*, 2010.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Learning as a functional state”***, José M. Delgado-García, plenary lecture, 4<sup>th</sup> Meeting of the Consorcio de Neurofisiología Clínica, Palma de Mallorca, Spain, 2011

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Neurophysiology of decision making”***, José M. Delgado-García, *International Workshop on Neurosciences and the Criminal Law*, Toledo, September, 2011.

Partner responsible: P05 UPO

Type of audience: Scientific community and experts in legal issues

***“Regenerative and compensatory mechanisms following peripheral and central nervous system lesions”***, José M. Delgado-García, plenary lecture, *International Workshop on Brain Reorganization after Spinal Cord Injury*, Toledo, May, 2011.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Learning and decisions as functional states of cortical circuits”***, José M. Delgado-García, *3rd International Conference on Cognitive Neurodynamics*, Niseko, Japan, June, 2011.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Learning as a functional state of the brain: studies in wild type and transgenic mice”***, José M. Delgado-García, *International symposium on Learning, Memory and Cognitive function*, Valencia, December, 2011.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Functional basis of associative learning: lessons from genetically manipulated animals”***, Agnès Gruart, Barcelona (Spain): *5<sup>th</sup> European Molecular and Cellular Cognition Society (EMCCS) - Federation of European Neuroscience Societies (FENS)*, 2012.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“The brain as a learning machine”***, José M. Delgado-García, *International Symposium on Biology and Communications*, Madrid, 26-27 March, 2012.

Partner responsible: P05 UPO

Type of audience: Scientific community and general audience

***“Red nucleus neurons actively contribute to the acquisition of classically conditioned eyelid responses in rabbits”***, José M. Delgado-García. *Neural Control of Movement Meeting*, Venice, Italy April, 22-23, 2012.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Learning as a functional state of the brain: studies in wild-type and transgenic animals”***, José M. Delgado-García, *Plenary lecture, 8th FENS Meeting*, Barcelona, Spain, July 16, 2012.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Red nucleus neurons actively contribute to the acquisition of an associative learning task in behaving rabbits”***, José M. Delgado-García, Carmona (Sevilla): *Dynamic Brain Platform(DBP)*, 2012.

Partner responsible: P05 UPO

Type of audience: Scientific community

***“Learning and decisions as functional states of cortical circuits”***, Agnès Gruart, Carmona (Sevilla): *Dynamic Brain Platform(DBP)*, 2012.

Partner responsible: P05 UPO  
Type of audience: Scientific community

***“Learning as a functional state: studies in wild-type and transgenic mice”***, José M. Delgado-García, Plenary lecture, 1st Meeting of the Federation of Latin-American Neuroscience Societies, Cancun, México, November, 2012.

Partner responsible: P05 UPO  
Type of audience: Scientific community

## **A4). PhD and Master theses**

### **A4.1). PhD thesis defense**

***“Genetic study of the heterogeneity of raphe serotonergic neurons”***

Vera Kiyasova, supervised by Patricia Gaspar, Paris (France), 06.09.2012.

Partner responsible: C001 INSERM  
Type of audience: Scientific community.

***“Conséquences comportementales et neurobiologiques d’une exposition précoce à l’alcool chez des souris consanguines”***

Jenny MOLET - Paris Descartes University (December 2010)

Partner responsible: C001b INSERM  
Type of audience: Scientific community.

***“Dissecting behavioral patterns and their neural circuits involved in fear and anxiety”***

Apar Jain, supervised by Cornelius Gross, Joint PhD award with University of Heidelberg, Germany. 17 September 2010.

Partner responsible: P02 EMBL  
Type of audience: Scientific community.

***“Novel Roles for the GABA<sub>B</sub> receptor in anxiety and cocaine addiction” (to be submitted December 2012)***, Fabian Sweeney.

Partner responsible: P04 UCC  
Type of audience: Scientific community.

***“Role of GABA<sub>B</sub> receptors in Stress, Anxiety and Depression” (to be submitted November 2012)***, Daniela Felice MSc

Partner responsible: P04 UCC  
Type of audience: Scientific community.

***“Hippocampal synaptic mechanisms underlying associative learning”***

Noelia Madroñal Monge, Supervised by José María Delgado-García and Agnès Gruart, Seville (Spain), 18.06.2010.

Partner responsible: P05 UPO  
Type of audience: Scientific community.



## A4.2). Dr. med thesis defense

***“Functional characterization of transgenic tissue-specific expression in mouse transactivator lines TPH2-tTA and mGFAP-tTA”***,

Max Rudolf Bauer, supervised by Tillmann Weber and Dusan Bartsch, Mannheim (Germany), 2011.

Partner responsible: P06 CIMH

Type of audience: Scientific community.

***“Tet-regulated inducible expression of candidate genes in serotonergic neurons of transgenic mice”***, Verena Karola Huppert, supervised by Tillmann Weber and Dusan Bartsch, Mannheim (Germany), 2011

Partner responsible: P06 CIMH

Type of audience: Scientific community.

## A4.3). Master thesis defense

Master 1: ***“Developmental role of SERT”***

Esmee Van Kleef, September 2011

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

Master 1: ***“Learning deficiencies in hyposerotoninergic mice”***

Elena Olive was supervised by Sebastian Fernandez- 06.2012

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

Master 1: ***“Development of serotonin ascending projections”***

Fanny Ledonne Master 1 defence supervised by Patricia Gaspar- 06.2012

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

Master 2: ***“Role of serotonin in the regulation of olfactory bulb neurogenesis***

Master 2 defence Florence Allain supervised by Patricia Gaspar and Sophie Scotto, Paris (France), 28.06.2012.

Partner responsible: CO01 INSERM

Type of audience: Scientific community.

***“Yif1B and 5-HT1A Receptors; A Characterisation of Yif1B -/- Mice A Novel Target for Antidepressants?”***

Brankele Frank – UPMC (June 2011)

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***“Do early antidepressant treatments produce changes in accumbal and midbrain serotonergic transmission in relation with impulsivity? Relevance to suicide”***

Aurélie Dahan – UPMC (June 2011)

Partner responsible: CO01b INSERM



Type of audience: Scientific community.

***"Is TrkB signaling underlying the behavioral and neurochemical effects of the novel antidepressant Agomelatine in the GR-1 murine model of depression?"***

Hester Velthuis Paris Descartes University (December 2011)

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"Implication du complexe bdnf/trkb dans l'effet de l'agomelatine dans un modèle de stress chronique chez la souris"***

Lara Zaidan – UPMC (June 2012)

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"Effet de l'inactivation du récepteur 5-HT3 sur la réponse aux antidépresseurs chez la souris"***

Vincent Martin - Paris Sud University (June 2012)

Partner responsible: CO01b INSERM

Type of audience: Scientific community.

***"An investigation into the neural circuits underlying the effects of a GABAB receptor positive modulator"***

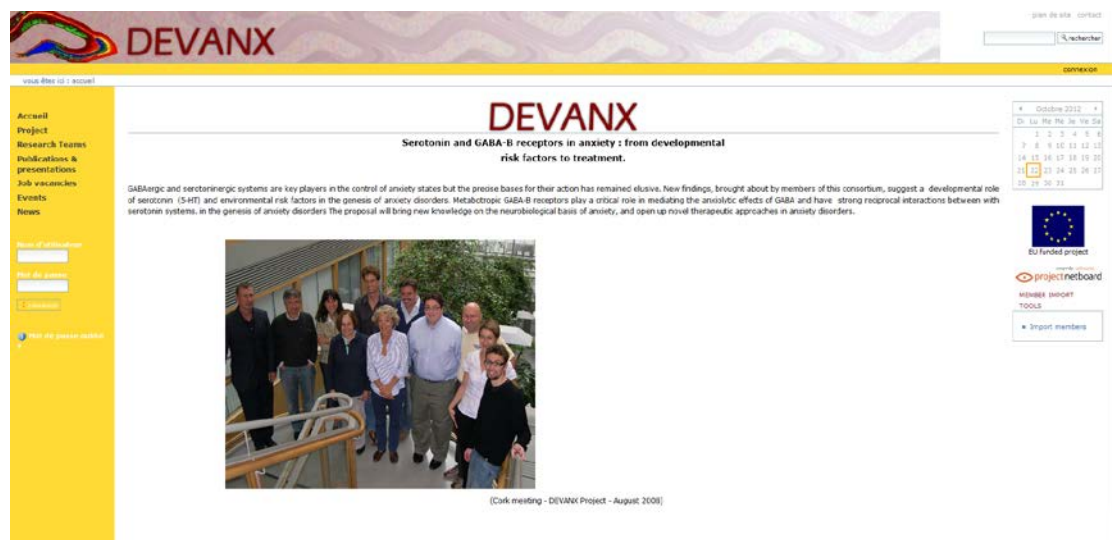
Riccardo Pizzo (2011)

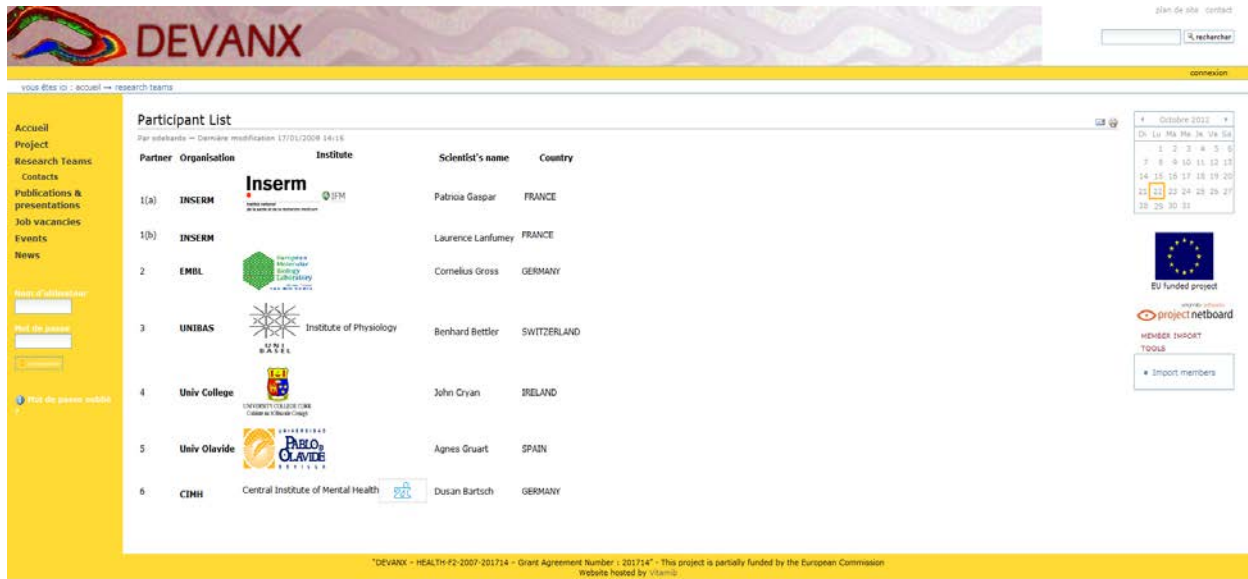
Partner responsible: P04 UCC

Type of audience: Scientific community.

## A5). Website

- Designed and managed by INSERM since the beginning of the DEVANX project.
- Allows an external visibility of the Project to anybody
- Can be reached at <http://devanx.vitamib.com/>
- Upgraded all along the project and a web link towards other web pages.





**Participant List**

2007-2012 - Dernière modification : 17/01/2008 14:16

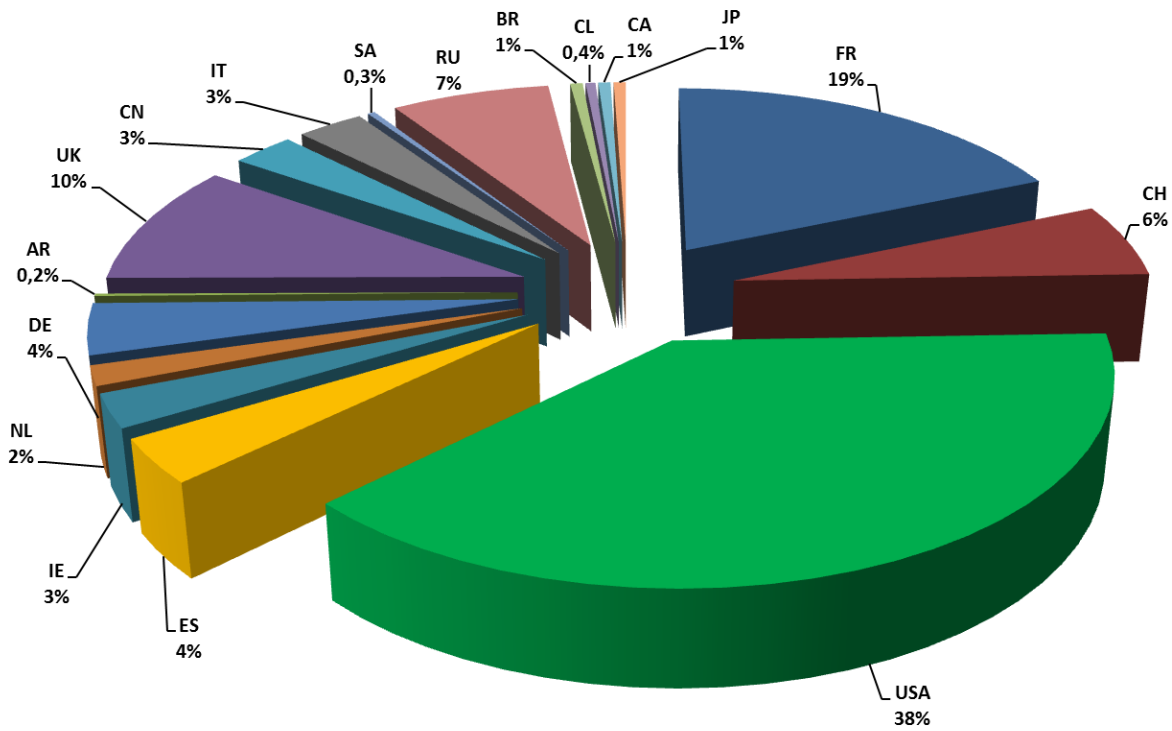
Partner	Organisation	Institute	Scientist's name	Country
1(a)	INSERM	Inserm	Patricia Gaspar	FRANCE
1(b)	INSERM		Laurence Lanfume	FRANCE
2	EMBL	European Molecular Biology Laboratory	Cornelius Gross	GERMANY
3	UNIBAS	Institute of Physiology	Bernhard Bettler	SWITZERLAND
4	Univ College	University College London	John Cryan	IRELAND
5	Univ Olavide	UNIVERSIDAD DE MADRID	Agnes Gruart	SPAIN
6	CIMH	Central Institute of Mental Health	Dusan Bartsch	GERMANY

DEVANX - HEALTH-F2-2007-201714 - Grant Agreement Number : 201714 - This project is partially funded by the European Commission  
Website hosted by : vitamed

## Website statistics

Period 2008-2012	
Visits	13036
Unique visitors	5775
# Countries	17
Average time/visit	126,2 sec
Average pages /visit	5,21

## Website visits broken down by countries



## SECTION B (CONFIDENTIAL)

### B) CONFIDENTIAL OR RESTRICTED PROJECT OUTPUTS

#### B1). Patents, Trademarks, Registered Designs, etc

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List of applications for patents, trademarks, registered designs, etc.			
Type of IP Rights: Patents, Trademarks, Registered designs, Utility models, etc.	Application reference(s) (e.g. EP123456)	Subject or title of application	Applicant (s) (as on the application)
None			

## B2). Exploitable foreground

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Overview table with exploitable foreground					
Exploitable Foreground (description)	Exploitable product(s) or measure(s)	Sector(s) of application	Timetable, commercial use	Patents or other IPR exploitation (licenses)	Owner and Other Beneficiary(s) involved
None					

### **B3). Description of exploitable foreground**

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Not applicable

### **B4). Consortium meetings**

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**DEVANX Kick Off Meeting**, Institut du Fer à Moulin, Paris (France), 05 December 2007.

Partner responsible: All

Type of meeting: consortium meeting

**DEVANX first report Meeting**, Cork (Ireland), 25 August 2008.

Partner responsible: All

Type of meeting: consortium

**DEVANX Consortium meetin**, Rome (Monterotondo, Italy), 22-23 January 2009.

Partner responsible: All

Type of meeting: consortium meeting

**DEVANX Consortium meeting**, Seville (Carmona, Spain), 12 March 2010.

Partner responsible: All

Type of meeting: consortium meeting

**DEVANX Consortium meeting**, Basel (Switzerland), 13 December 2010.

Partner responsible: UNIBASEL (Benny Bettler)

Type of meeting: consortium meeting

**DEVANX Consortium meeting**, Mannheim (Germany), 13 January 2012.

Partner responsible: CIMH (Dusan Bartsch)

Type of meeting: consortium meeting

**DEVANX Conclusion Meeting and Symposium**,

Institut du Fer à Moulin, Paris (France), 17-18 June 2012.

Partner responsible: All

Type of audience: Scientific community. A one day scientific meeting was organized and was open to the community.



### 3 Report on societal implications

#### A General Information *(completed automatically when Grant Agreement number is entered).*

Grant Agreement Number:	201714
Title of Project:	"Serotonin and GABA-B receptors in anxiety: from developmental risk factors to treatment"
Name and Title of Coordinator:	Dr Patricia Gaspar

#### B Ethics

<b>1. Did your project undergo an Ethics Review (and/or Screening)?</b>	<input type="radio"/> Yes <input type="radio"/> No
<ul style="list-style-type: none"> <li>If Yes: have you described the progress of compliance with the relevant Ethics Review/Screening Requirements in the frame of the periodic/final project reports?</li> </ul> <p>Special Reminder: the progress of compliance with the Ethics Review/Screening Requirements should be described in the Period/Final Project Reports under the Section 3.2.2 'Work Progress and Achievements'</p>	
<b>2. Please indicate whether your project involved any of the following issues (tick box) :</b>	<b>YES</b>
<b>RESEARCH ON HUMANS</b>	
• Did the project involve children?	NO
• Did the project involve patients?	NO
• Did the project involve persons not able to give consent?	NO
• Did the project involve adult healthy volunteers?	NO
• Did the project involve Human genetic material?	NO
• Did the project involve Human biological samples?	NO
• Did the project involve Human data collection?	NO
<b>RESEARCH ON HUMAN EMBRYO/FOETUS</b>	
• Did the project involve Human Embryos?	NO
• Did the project involve Human Foetal Tissue / Cells?	NO
• Did the project involve Human Embryonic Stem Cells (hESCs)?	NO
• Did the project on human Embryonic Stem Cells involve cells in culture?	NO
• Did the project on human Embryonic Stem Cells involve the derivation of cells from Embryos?	NO
<b>PRIVACY</b>	
• Did the project involve processing of genetic information or personal data (eg. health, sexual lifestyle, ethnicity, political opinion, religious or philosophical conviction)?	NO
• Did the project involve tracking the location or observation of people?	NO
<b>RESEARCH ON ANIMALS</b>	
• Did the project involve research on animals?	YES
• Were those animals transgenic small laboratory animals?	YES
• Were those animals transgenic farm animals?	NO
• Were those animals cloned farm animals?	NO

• Were those animals non-human primates?	NO
<b>RESEARCH INVOLVING DEVELOPING COUNTRIES</b>	
• Did the project involve the use of local resources (genetic, animal, plant etc)?	NO
• Was the project of benefit to local community (capacity building, access to healthcare, education etc)?	NO
<b>DUAL USE</b>	
• Research having direct military use	NO
• Research having the potential for terrorist abuse	NO

## C Workforce Statistics

**3. Workforce statistics for the project: Please indicate in the table below the number of people who worked on the project (on a headcount basis).**

Type of Position	Number of Women	Number of Men
Scientific Coordinator	1	0
WP leaders	1	7
Experienced researchers	12	15
PhD Students	13	17
Others	1	0
<b>4. How many additional researchers (in companies and universities) were recruited specifically for this project?</b>		7
Of which, indicate the number of men: Senior Researchers ; Research Fellows ; PhD students ; Young Researchers		5

<b>D Gender Aspects</b>		
<b>5. Did you carry out specific Gender Equality Actions under the project?</b>	<input type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No
<b>6. Which of the following actions did you carry out and how effective were they?</b>		
	Not at all effective	Very effective
<input type="checkbox"/> Design and implement an equal opportunity policy	○ ○ ○ ○ ○	○ ○ ○ ○ ○
<input type="checkbox"/> Set targets to achieve a gender balance in the workforce	○ ○ ○ ○ ○	○ ○ ○ ○ ○
<input type="checkbox"/> Organise conferences and workshops on gender	○ ○ ○ ○ ○	○ ○ ○ ○ ○
<input type="checkbox"/> Actions to improve work-life balance	○ ○ ○ ○ ○	○ ○ ○ ○ ○
<input type="radio"/> Other: <span style="border: 1px solid black; padding: 2px 10px;">NA</span>		
<b>7. Was there a gender dimension associated with the research content – i.e. wherever people were the focus of the research as, for example, consumers, users, patients or in trials, was the issue of gender considered and addressed?</b>		
<input type="radio"/> Yes- please specify <span style="border: 1px solid black; display: inline-block; width: 150px; height: 20px; vertical-align: middle;"></span>		
<input checked="" type="radio"/> No		
<b>E Synergies with Science Education</b>		
<b>8. Did your project involve working with students and/or school pupils (e.g. open days, participation in science festivals and events, prizes/competitions or joint projects)?</b>		
<input checked="" type="radio"/> Yes- please specify: brain awareness week; hosting high school students.		
<input type="radio"/> No		
<b>9. Did the project generate any science education material (e.g. kits, websites, explanatory booklets, DVDs)?</b>		
<input type="radio"/> Yes- please specify <span style="border: 1px solid black; display: inline-block; width: 150px; height: 20px; vertical-align: middle;"></span>		
<input checked="" type="radio"/> No		
<b>F Interdisciplinarity</b>		
<b>10. Which disciplines (see list below are involved in your project?</b>		
3.1 <input type="radio"/> Main discipline <sup>3</sup> :		
1.5 <input type="radio"/> Associated discipline <sup>3</sup> :	<input type="radio"/> Associated discipline <sup>3</sup> : 5.1	
<b>G Engaging with Civil society and policy makers</b>		
<b>11a Did your project engage with societal actors beyond the research community? (if 'No', go to Question 14)</b>	<input type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No

<sup>3</sup> Insert number from list below (Frascati Manual).

<b>11b If yes, did you engage with citizens (citizens' panels / juries) or organised civil society (NGOs, patients' groups etc.)?</b>					
<input type="radio"/> No <input type="radio"/> Yes- in determining what research should be performed <input type="radio"/> Yes - in implementing the research <input type="radio"/> Yes, in communicating /disseminating / using the results of the project					
<b>11c In doing so, did your project involve actors whose role is mainly to organise the dialogue with citizens and organised civil society (e.g. professional mediator; communication company, science museums)?</b>				<input type="radio"/> <input type="radio"/>	Yes No
<b>12. Did you engage with government / public bodies or policy makers (including international organisations)</b>					
<input type="radio"/> No <input type="radio"/> Yes- in framing the research agenda <input type="radio"/> Yes - in implementing the research agenda <input type="radio"/> Yes, in communicating /disseminating / using the results of the project					
<b>13a Will the project generate outputs (expertise or scientific advice) which could be used by policy makers?</b>					
<input type="radio"/> Yes – as a <b>primary</b> objective (please indicate areas below- multiple answers possible) <input type="radio"/> Yes – as a <b>secondary</b> objective (please indicate areas below - multiple answer possible) <input type="radio"/> No					
<b>13b If Yes, in which fields?</b>					
Agriculture Audiovisual and Media Budget Competition Consumers Culture Customs Development Economic and Monetary Affairs Education, Training, Youth Employment and Social Affairs	Energy Enlargement Enterprise Environment External Relations External Trade Fisheries and Maritime Affairs Food Safety Foreign and Security Policy Fraud Humanitarian aid	Human rights Information Society Institutional affairs Internal Market Justice, freedom and security Public Health Regional Policy Research and Innovation Space Taxation Transport			

**13c If Yes, at which level?**

- ☐ Local / regional levels  
☐ National level  
☐ European level  
☐ International level

**H Use and dissemination**
**14. How many Articles were published/accepted for publication in peer-reviewed journals?**
**73**
**To how many of these is open access<sup>4</sup> provided?**
**55**
**How many of these are published in open access journals?**
**55**
**How many of these are published in open repositories?**
**0**
**To how many of these is open access not provided?**
**18**
**Please check all applicable reasons for not providing open access:**

- ☐ publisher's licensing agreement would not permit publishing in a repository  
☐ no suitable repository available  
☐ no suitable open access journal available  
☐ no funds available to publish in an open access journal  
☐ lack of time and resources  
☒ lack of information on open access  
☐ other<sup>5</sup>: .....

**15. How many new patent applications ('priority filings') have been made?** (*"Technologically unique": multiple applications for the same invention in different jurisdictions should be counted as just one application of grant*).

**None**
**16. Indicate how many of the following Intellectual Property Rights were applied for (give number in each box).**

Trademark

**None**

Registered design

**None**

Other

**None**
**17. How many spin-off companies were created / are planned as a direct result of the project?**
**None**
*Indicate the approximate number of additional jobs in these companies:*
**None**
**18. Please indicate whether your project has a potential impact on employment, in comparison with the situation before your project:**

- |   |   |
|---|---|
| <input type="checkbox"/> Increase in employment, or                       | <input type="checkbox"/> In small & medium-sized enterprises                        |
| <input type="checkbox"/> Safeguard employment, or                         | <input type="checkbox"/> In large companies   |
| <input type="checkbox"/> Decrease in employment,                          | <input checked="" type="checkbox"/> None of the above / not relevant to the project |
| <input type="checkbox"/> Difficult to estimate / not possible to quantify |   |

<sup>4</sup> Open Access is defined as free of charge access for anyone via Internet.

<sup>5</sup> For instance: classification for security project.

<b>19. For your project partnership please estimate the employment effect resulting directly from your participation in Full Time Equivalent (FTE = one person working fulltime for a year) jobs:</b>		Indicate figure:  <b>0</b>
Difficult to estimate / not possible to quantify		<input type="checkbox"/>
<b>I Media and Communication to the general public</b>		
<b>20. As part of the project, were any of the beneficiaries professionals in communication or media relations?</b> <input type="radio"/> Yes <input checked="" type="radio"/> No		
<b>21. As part of the project, have any beneficiaries received professional media / communication training / advice to improve communication with the general public?</b> <input type="radio"/> Yes <input checked="" type="radio"/> No		
<b>22 Which of the following have been used to communicate information about your project to the general public, or have resulted from your project?</b>		
<input checked="" type="checkbox"/> Press Release <input checked="" type="checkbox"/> Media briefing <input checked="" type="checkbox"/> TV coverage / report <input checked="" type="checkbox"/> Radio coverage / report <input checked="" type="checkbox"/> Brochures /posters / flyers <input type="checkbox"/> DVD /Film /Multimedia	<input checked="" type="checkbox"/> Coverage in specialist press <input type="checkbox"/> Coverage in general (non-specialist) press <input checked="" type="checkbox"/> Coverage in national press <input checked="" type="checkbox"/> Coverage in international press <input checked="" type="checkbox"/> Website for the general public / internet <input checked="" type="checkbox"/> Event targeting general public (festival, conference, exhibition, science café)	
<b>23 In which languages are the information products for the general public produced?</b>		
<input type="checkbox"/> Language of the coordinator <input type="checkbox"/> Other language(s)	<input checked="" type="checkbox"/> English	

**Question F-10:** Classification of Scientific Disciplines according to the Frascati Manual 2002 (Proposed Standard Practice for Surveys on Research and Experimental Development, OECD 2002):

#### **FIELDS OF SCIENCE AND TECHNOLOGY**

##### 1. NATURAL SCIENCES

- 1.1 Mathematics and computer sciences [mathematics and other allied fields: computer sciences and other allied subjects (software development only; hardware development should be classified in the engineering fields)]
- 1.2 Physical sciences (astronomy and space sciences, physics and other allied subjects)
- 1.3 Chemical sciences (chemistry, other allied subjects)
- 1.4 Earth and related environmental sciences (geology, geophysics, mineralogy, physical geography and other geosciences, meteorology and other atmospheric sciences including climatic research, oceanography, vulcanology, palaeoecology, other allied sciences)
- 1.5 Biological sciences (biology, botany, bacteriology, microbiology, zoology, entomology, genetics, biochemistry, biophysics, other allied sciences, excluding clinical and veterinary sciences)

##### 2. ENGINEERING AND TECHNOLOGY

- 2.1 Civil engineering (architecture engineering, building science and engineering, construction engineering, municipal and structural engineering and other allied subjects)



- 2.2 Electrical engineering, electronics [electrical engineering, electronics, communication engineering and systems, computer engineering (hardware only) and other allied subjects]
- 2.3. Other engineering sciences (such as chemical, aeronautical and space, mechanical, metallurgical and materials engineering, and their specialised subdivisions; forest products; applied sciences such as geodesy, industrial chemistry, etc.; the science and technology of food production; specialised technologies of interdisciplinary fields, e.g. systems analysis, metallurgy, mining, textile technology and other applied subjects)
- 3. MEDICAL SCIENCES
  - 3.1 Basic medicine (anatomy, cytology, physiology, genetics, pharmacy, pharmacology, toxicology, immunology and immunohaematology, clinical chemistry, clinical microbiology, pathology)
  - 3.2 Clinical medicine (anaesthesiology, paediatrics, obstetrics and gynaecology, internal medicine, surgery, dentistry, neurology, psychiatry, radiology, therapeutics, otorhinolaryngology, ophthalmology)
  - 3.3 Health sciences (public health services, social medicine, hygiene, nursing, epidemiology)
- 4. AGRICULTURAL SCIENCES
  - 4.1 Agriculture, forestry, fisheries and allied sciences (agronomy, animal husbandry, fisheries, forestry, horticulture, other allied subjects)
  - 4.2 Veterinary medicine
- 5. SOCIAL SCIENCES
  - 5.1 Psychology
  - 5.2 Economics
  - 5.3 Educational sciences (education and training and other allied subjects)
  - 5.4 Other social sciences [anthropology (social and cultural) and ethnology, demography, geography (human, economic and social), town and country planning, management, law, linguistics, political sciences, sociology, organisation and methods, miscellaneous social sciences and interdisciplinary , methodological and historical S1T activities relating to subjects in this group. Physical anthropology, physical geography and psychophysiology should normally be classified with the natural sciences].
- 6. HUMANITIES
  - 6.1 History (history, prehistory and history, together with auxiliary historical disciplines such as archaeology, numismatics, palaeography, genealogy, etc.)
  - 6.2 Languages and literature (ancient and modern)
  - 6.3 Other humanities [philosophy (including the history of science and technology) arts, history of art, art criticism, painting, sculpture, musicology, dramatic art excluding artistic "research" of any kind, religion, theology, other fields and subjects pertaining to the humanities, methodological, historical and other S1T activities relating to the subjects in this group]