RAPID ANXIOLYTIC EFFECTS OF 5-HT$_4$ RECEPTOR AGONIST INVOLVES PREFRONTAL CORTEX-BRAINSTEM NEURAL CIRCUIT RECRUITMENT

Faye C.[1], Hen R.[2], Guiard B.[3], Denny C.A.[2], Gardier A.M.[1], Mendez-David I.[1,*], David D.J.[1,*,§]

[1]CESP/UMRS1178, Univ Paris-Sud, Fac Pharmacie, INSERM, Université Paris-Saclay, Châtenay-Malabry, France;
[2]Department of Psychiatry, Columbia University, NYC, USA; Division of Integrative Neuroscience, New York State Psychiatric Institute, NYC, USA;
[3]UMR5169 CNRS "Centre de Recherches sur la Cognition Animale", Toulouse, France

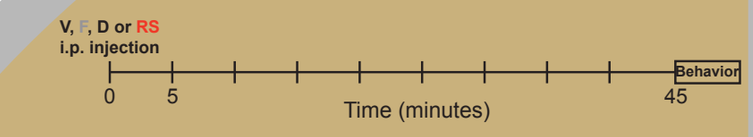
INTRODUCTION:

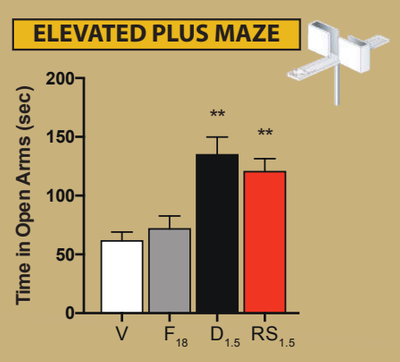
Benzodiazepines are an effective treatment of anxiety disorders for years, but misgiving over side effects have led to prescribe some antidepressant drugs even though they present a delayed onset of action[1].

AIM:

Here, we sought to evaluate whether serotonin type 4 receptor (5-HT4R) activation and targeting glutamatergic axon terminals arising from the medial prefrontal cortex (mPFC) in the dorsal raphe nucleus (DRN) may constitute a new way to induce fast anxiolytic effects[2].

1 SYSTEMIC and ACUTE 5-HT$_4$R stimulation induced FAST anxiolytic effect



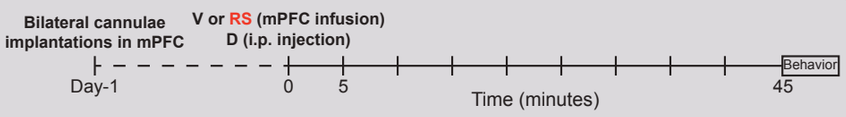


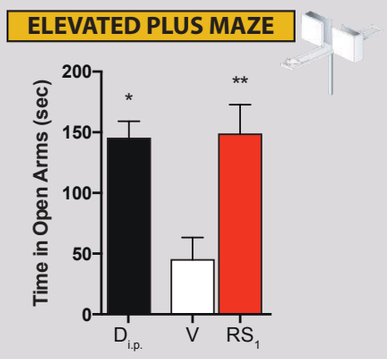
**p<0.01 vs vehicle

	Fluoxetine	Diazepam	RS67333
ELEVATED PLUS MAZE	Ø	+	+
OPEN FIELD	Ø	+	+
NOVELTY SUPPRESSED FEEDING	Ø	+	+

Abreviations: D$_{1.5}$: diazepam 1.5mg/kg; F$_{18}$: fluoxetine 18mg/kg; RS$_{1.5}$: RS67333 1.5mg/kg; V: vehicle 0.9%; Ø: no effect; +: anxiolytic-like effect

2 LOCAL and ACUTE 5-HT$_4$R stimulation induced FAST anxiolytic effect

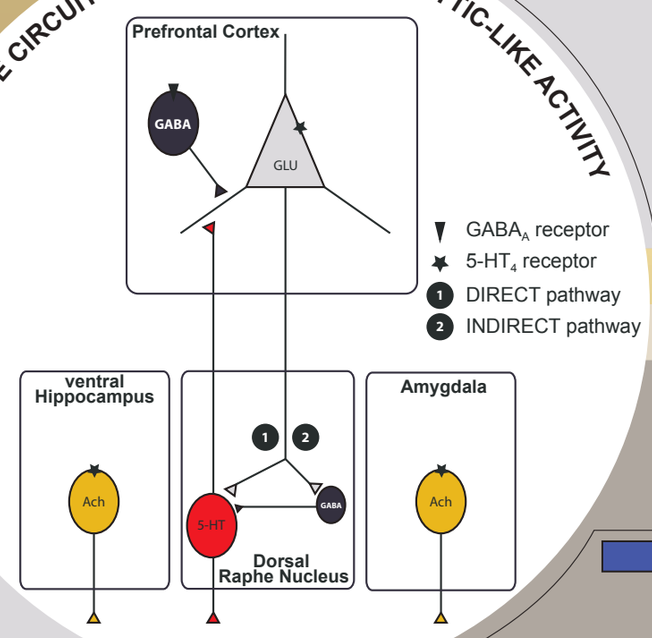




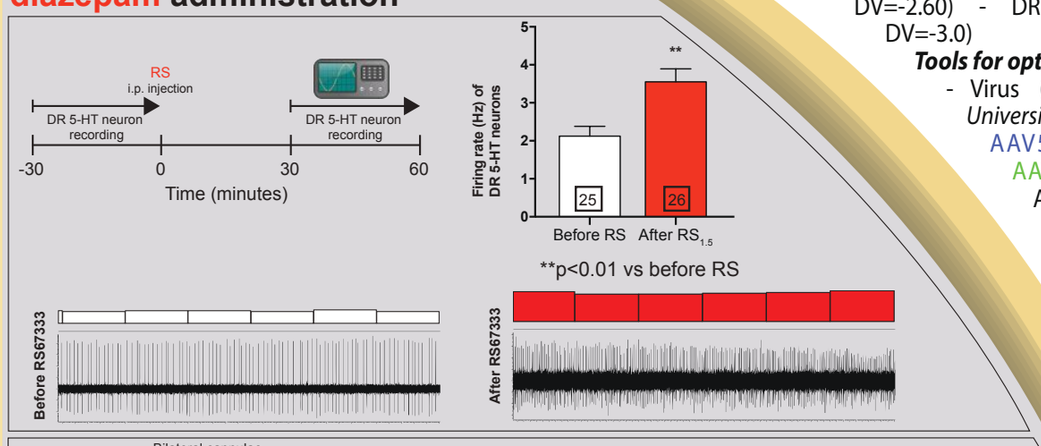
*p<0.05; **p<0.01 vs vehicle

	Diazepam	RS67333
ELEVATED PLUS MAZE	+	+
NOVELTY SUPPRESSED FEEDING	+	+

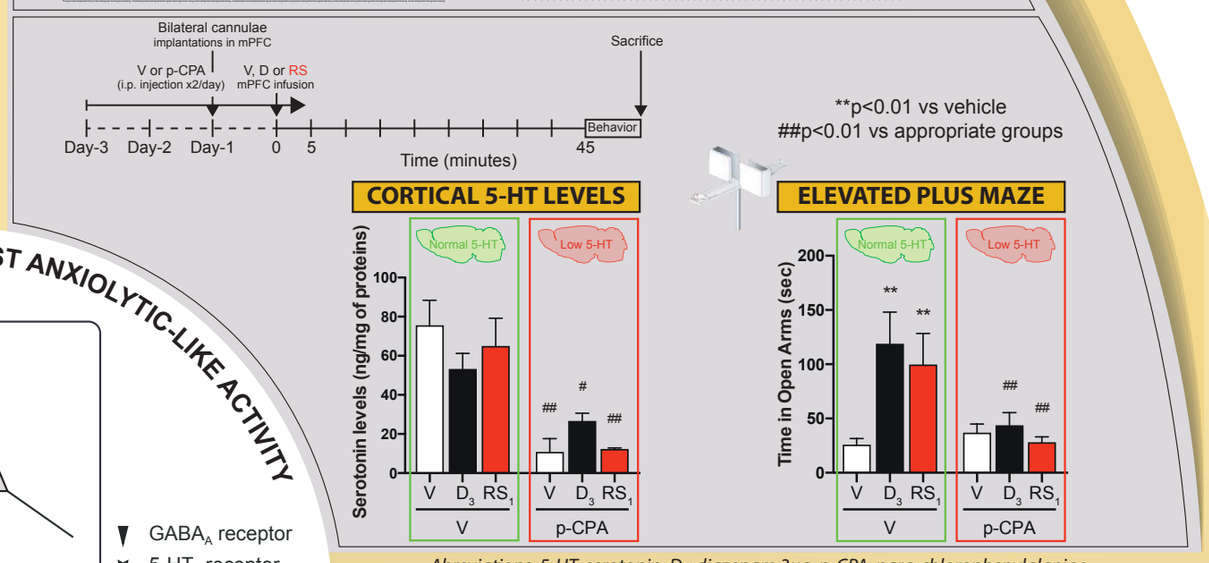
PUTATIVE CIRCUIT INVOLVED IN FAST ANXIOLYTIC-LIKE ACTIVITY



3 DRN 5-HT is involved in FAST anxiolytic effect of acute RS67333 and diazepam administration[4]



**p<0.01 vs before RS



**p<0.01 vs vehicle
##p<0.01 vs appropriate groups

Abreviations: 5-HT: serotonin; D$_3$: diazepam 3µg; p-CPA: para-chlorophenylalanine RS$_{1.5}$: RS67333 1.5mg/kg; RS$_1$: RS67333 1µg; V: vehicle 0.9%

MATERIEL AND METHODS:

Animals: male BALB/cJRj mice, 8 weeks (20-25g), with anxiety-like phenotype at basal state[3]

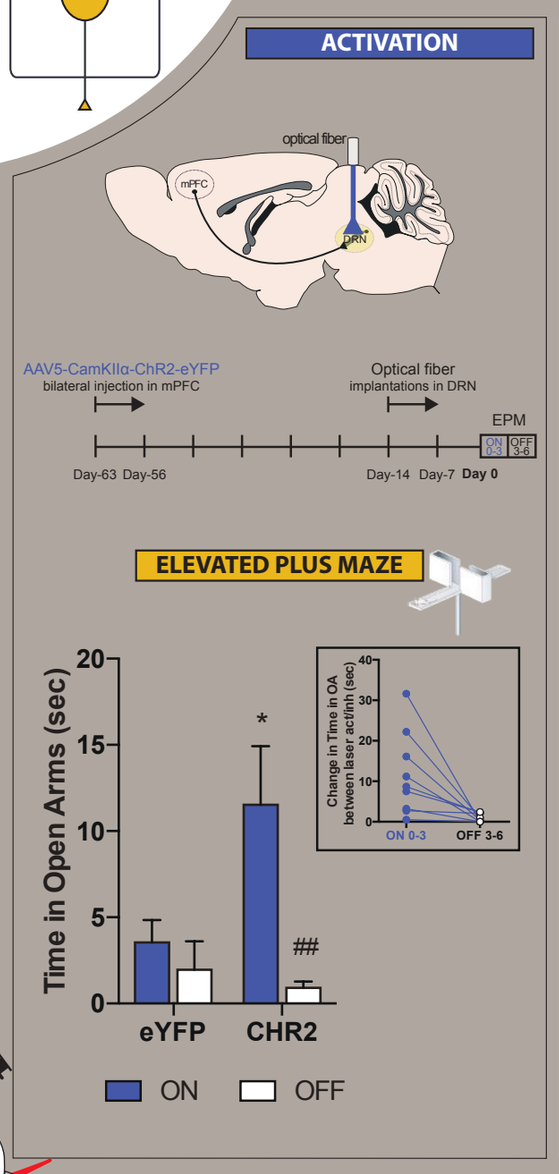
Drugs: NaCl (vehicle): 0.9% - fluoxetine (SSRI, Anawa Trading, Switzerland): 18mg/kg - diazepam (benzodiazepine, Sigma-Aldrich, France): 1.5mg/kg or 3µg - RS67333 (5-HT$_4$ agonist, pKi=8.7): 1.5mg/kg, 0.2µg or 1µg (Tocris, UK) - para-chlorophenylalanine (p-CPA, tryptophan hydroxylase inhibitor, Sigma-Aldrich, France): 300mg/kg/day

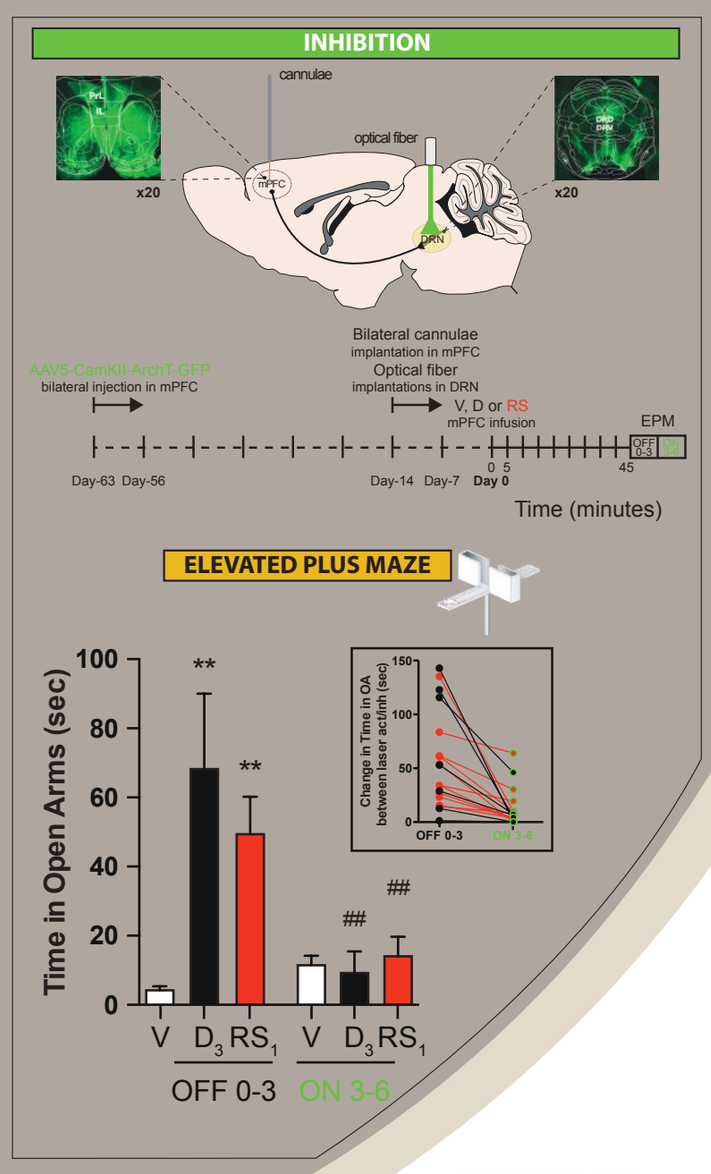
Behavioral paradigms: Elevated Plus maze (EPM) - Novelty Suppressed Feeding (NSF) - Open Field (OF)

Stereotaxic coordinates (in mm relative to Bregma): mPFC (AP=+2.10; LL=±0.50; DV=-2.60) - DRN (AP=-4.50; LL=1.0; DV=-3.0)

Tools for optogenetic:
- Virus (*from Ed Boyden, MIT University, USA*): AAV5-CamKII-ChR2-eYFP, AAV5-CamKII-ArchT-GFP, AAV5-CamKII-GFP
- 473nm blue light, 10ms pulses, 10Hz, 10mW, 3min
- 532nm green light, 15mW, 3min

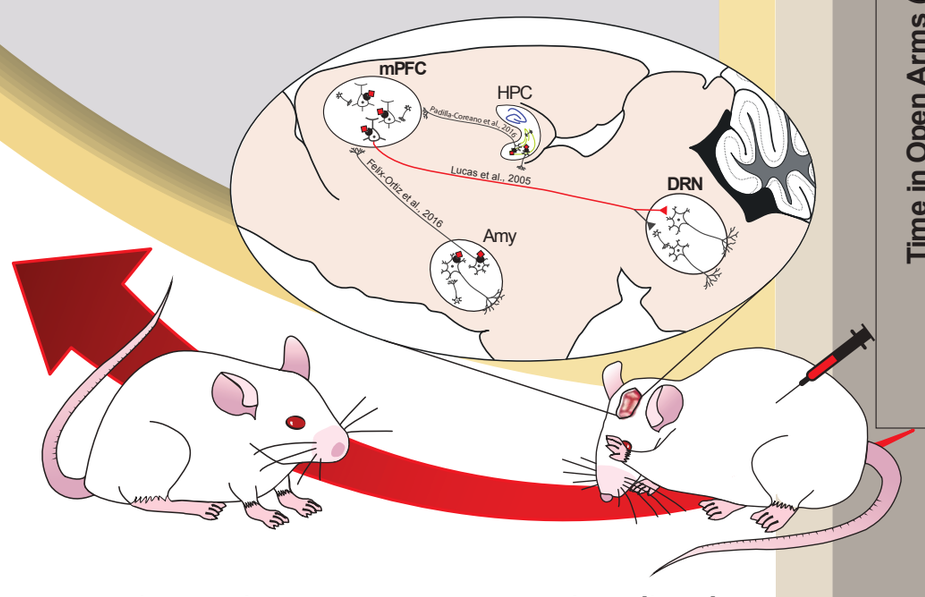
4 mPFC GLUTAMATERGIC TERMINALS into DRN contributed to FAST anxiolytic effects related to 5-HT$_4$R stimulation[5]





*p<0.05 vs eYFP-LASER ON
##p<0.01 vs CHR2-LASER ON

**p<0.01 vs vehicle
##p<0.01 vs appropriate groups (LASER OFF)



CONCLUSION:

1- 5-HT$_4$R agonist induces fast anxiolytic effect in anxious mice similar to classical benzodiazepine .

2- mPFC contributes to the fast anxiolytic effect induced by RS67333 and diazepam.

3- DRN 5-HT synthesis is essential for cortical RS67333 and diazepam activity.

4- Cortical glutamatergic terminals in the DRN mediate the fast anxiolytic effect induced by RS67333 and diazepam.

5-HT$_4$R stimulation could represent an innovative strategy to treat anxiety disorders with similar anxiolytic potential as benzodiazepine. Despite different pharmacological targets, RS67333 and diazepam, share common mechanisms to induced fast anxiolytic effect through mPFC-brainstem circuit recruitment.

REFERENCES:

[1]Samuels et al., 2016, The Neuroscientist
[2]Mendez-David et al., 2014, NPP
[3]Dulawa et al., 2004, NPP
[4]Lucas et al., 2004, Biol Psy
[5]Warden et al., 2012, Nature

*NO CONFLICT OF INTEREST