TITLE:

The startle reflex, a measure of motivation, attention and psychopathology: parallel evolutionin humans and *drosophila melanogaster*

ABSTRACT (max 300 words):
The startle reflex is a fast defensive response aimed at avoiding sensory or physical injury and highly preserved in the animal kingdom. The reflex is elicited by rapid and intense sensory stimulation like a shot or a flash. The neural circuit involves only a few synapses and giant neurons: it starts within 3-10 ms from stimulation onset, is similar across all species, and consists of an intense muscular flexion response which involves the whole body. In humans, the main neural circuit of the acoustic startle is considered similar to the most studied circuit in the rat (Yeomans & Frankland, 1996) and is typically investigated by measuring the amplitude of muscular (EMG) contraction of the *orbicularis oculi* during the elicited eyeblink. Interestingly, the amplitude of the reflex is modulated by emotions, anxiety (Poli & Angrilli, 2015),attention, but it represents also an interesting probe which is impaired in psychiatric (e.g. schizophrenia) and neurological patients. Furthermore, the startle reflex is controlled by the amygdala (Angrilli et al., 1996) and the orbitofrontal cortex. In the *drosophila* it is measured through the action potential of the indirect flight muscle (Zordan et al., 2005) (Fig 1-2).Similarly to mammals, also in drosophila a giant fiber pathway is involved and two different avoidance defensive responses are possible: a fast imprecise and a slower more coordinated one (von Rein et al., 2014). Aim of the present project is to investigate the still unknown evolutionistic parallel between these two different startle systems by adapting and translating several paradigms developed from one species to the other. The project will allow to implement new approaches and tools useful for both basic research and possible applicationsto the diagnosis and treatments of neurological and psychiatric disorders.

REFERENCES (Max 5):

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2) Zordan, M.A., Massironi, M. Ducato, M.G., te Kronnie, G., Costa, R., Reggiani, C., Carine Chagneau, C., Martin, J. R. &Megighian, A. (2005). Drosophila CAKI/CMG Protein, a Homolog of Human CASK, Is Essential for Regulation of Neurotransmitter Vesicle Release.*Journal of Neurophysiology*, 94, 1074–1083.
3) Yeomans, J.S. &Frankland, P.W. (1996). The acoustic startle reflex: neurons and connections. *Brain ResearchReviews*, 21, 301-314.
4) Poli, E., Angrilli, A. (2015). Greater general startle reflex is associated with greater anxiety levels: a correlational study on 111 young women. *Frontiers in Behavioral Neuroscience*, 9, 1-6. doi: 10.3389/fnbeh.2015.00010
5) von Reyn, C.R, Breads, P., Peek, M.Y. Zheng, G.Z., Williamson, W.R., Yee, A.L., Leonardo, A. & Card, G.M. (2014). A spike-timing mechanism for action selection. *Nature Neuroscience*, 17, 962-970.

PARTICIPANTS (PI and co-PIs):
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Co-PI: Alessandro Angrilli

EXPERIMENTAL DATA:

To be acquired	X
Already acquired (ready to be used)	

If data need to be acquired, please provide a brief description of the Experimental setup, methods, instruments and scheduling (e.g. # of subjects, images/signals…): max 300 words

Four experiments will be carried out, two on humans and two on *drosophila*. In the first year of PhD, two parallel studies will be implemented on different species with the aim to investigate

similarities of startle response under appetitive, aversive and attentional conditions. In the first research on *drosophila*, startle reflex is administered in a randomly repeated series to 30 flies during appetitive stimulation (food presentation), aversive condition (quickly enlarging object presented through a monitor), and orienting to a point-wise light. In the parallel experiment on humans a sample of 30 students will be enrolled. Acoustic startle reflex will be randomly elicited during the presentation of a set of standardized emotional slides divided in pleasant, neutral and unpleasant, and in a second session during the Mackworth Clock Test, a test of sustained attention. It is expected that startle reflex is inhibited during pleasant stimulation and attentional load and potentiated during aversive stimulation.

The second couple of studies will be carried out in the second year of the PhD period and will focus on modulation of startle reflex amplitude by dopamine levels. It is expected greater reflex associated to higher levels of brain dopamine and vice versa. Thus, startle reflex and pre-pulse modulation will be investigated in both *drosophila* genetically modified for having low (with DTH mutation; Cichewicz et al., 2016) vs. high (amphetamine administration) levels of dopamine. Similarly two groups of students will be selected from a large sample for having low vs low levels of fronto-striatal dopamine. This can be accomplished through an index measured in resting state conditions, the eyeblink rate (Jongkees & Colzato, 2016), individuals with highest rate of blink rate have larger fronto-striatal dopamine. The experiments in humans and drosophila are compared at level of pattern analysis in which the different conditions are compared within each species and non parametric statistics will be used for directly comparing between-species experiments.

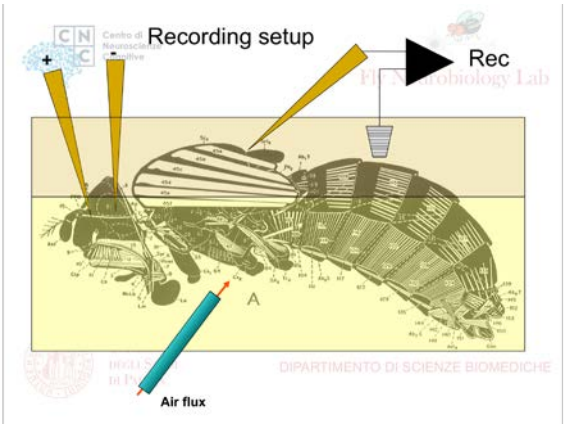

Fig 1: Recording setup for electrophysiologically measuring startle reflex in *D.melanogaster* (in this case brain stimulation).

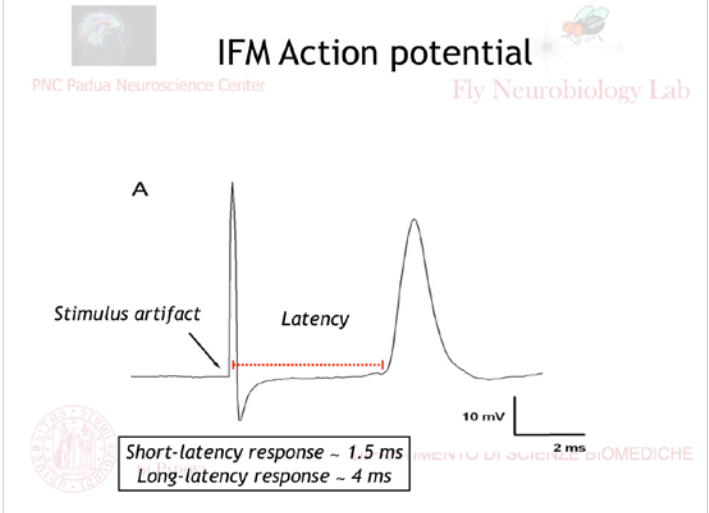

Fig 2. Intracellular recording of a long-latency action potential evoked in an Indirect Flight Muscle fibre by triggering startle reflex.

ETHICS COMMITTEE:

Obtained	
Conditioned submission*	Expected time response (in months): **2-3 months**
Not required	

* request will be submitted only if a PhD student will be associated to the project