# OREXIN SYSTEM: NETWORK MULTI-TASKING

RAFFAELE SPERANDEO[1,8], MAURO N. MALDONATO[2,8], ANTONIETTA MESSINA[2,9], PASQUALE COZZOLINO[3], MARCELLINO MONDA[2], FRANCESCO CERRONI[4], PALMIRA ROMANO[4], MARGHERITA SALERNO[6], AGATA MALTESE[6], MICHELE ROCCELLA[6], LUCIA PARISI[6], GABRIELE TRIPI[7], FIORENZO MOSCATELLI[8], FRANCESCO SESSA[8], MONICA SALERNO[8], GIUSEPPE CIBELLI[8], GIOVANNI MESSINA[8], VINCENZO MONDA[9], SERGIO CHIEFFI[9], INES VILLANO[9], EMANUELE MONDA[9], MARIA RUBERTO[10], GABRIELLA MARSALA[11], ROSA MAROTTA R[5], ANNA VALENZANO[8*]

[1]Department of Experimental Medicine, Section of Human Physiology and Unit of Dietetic and Sport Medicine, Università degli Studi della Campania, Naples, Italy - [2]Department of Human science, Università della Basilicata, Italy - [3]SiPGI Postgraduate School of Integrated Gestalt Psychotherapy, D.M. 12.10.2007 Naples, Italy - [4]Clinic of Child and Adolescent Neuropsychiatry, Department of Mental Health, Physical and Preventive Medicine, Università degli Studi della Campania "Luigi Vanvitelli", Italy - [5]Department of Health Sciences, University "Magna Graecia", Catanzaro, Italy - [6]Child Neuropsychiatry, Department of Psychology and Pedagogical Sciences, University of Palermo, Palermo, Italy - [7]Child Neuropsychiatry, Department of Psychology and Pedagogical Sciences, University of Palermo, Palermo, Italy - [8]Department of Clinical and Experimental Medicine, University of Foggia, Foggia, Italy [9]Department of Experimental Medicine, Section of Human Physiology and Unit of Dietetics and Sports Medicine, Università degli Studi della Campania "Luigi Vanvitelli", Naples, Italy - [10]Centro CRD, Santa Maria del Pozzo, Somma Vesuviana, Naples - [11]Struttura Complessa di Farmacia, Azienda Ospedaliero-Universitaria, Foggia, Italy

***ABSTRACT***

*Orexin system regulates sleep/wake states and its deficiency result in narcolepsy thus indicating the crucial role of orexins in maintaining wakefulness. There are two types of orexin peptides: the orexin-A (OXA or hypocretin 1) and orexin-B (OXB or hypocretin 2). The Majority of the central nervous system orexin peptides are synthesized in neurons located in the lateral and back hypothalamus and send projections throughout the brain regions Orexin neurons are "multi-tasking" hence regulating also energy homeostasis, reward systems and feeding behaviour through connection with hypothalamic nuclei and through responsiveness to leptine and glucose. It has recently been found a connection with lymbic system suggesting a further possible role of orexins in regulating emotions. All the studies conducted confirm that orexin system regulates vigilance states, energy homeostasis, reward system, and emotions. These crucial role might be the target to develope treatments of narcolepsy, obesity, emotional stress, and drug addiction.*

***Keywords****: Orexin, Obesity, emotional stress, narcolepsy.*

*DOI: 10.19193/0393-6384_2018_2_55*

***Received*** *November 30, 2017;* ***Accepted*** *January 20, 2018*

## Introduction

Orexin A and B are excitatory hypothalamic neuropeptides, which play a key role in various physiologic functions such as sleep and wakefullness regulation, thermoregulation, control of energy metabolism, cardiovascular responses, feeding, and SPA (spontaneous physical activity).

Described by Sakurai et al.[(1)] at the end of the last century (referred to as Hypocretins 1 and 2) they were firstly recognized as regulators of feeding behaviour because of their production in a hypothalamic region known as the feeding centre[(2-5)]; further research showed a clear association between hypocretins system and a sleep disorder named narcolepsy hence indicating a regulation of sleep and wakefulness[(5-9)]. More recent studies focused the attention on the role of the orexins in the coordination of emotion, energy homeostasis, reward, drug addiction and arousal[(10-14)]. These findings suggest that orexin/hypocretin system is involved in regulation of many different physiologic functions and that its implications in metabolism pathways are far to be definetely determined.

## The orexin/hypocretin system

Orexins are two neuropeptides synthesized, in humans as in mammals, in neurons of lateral hypothalamic and perifornical area[15-17]. The two subtypes of orexin (A and B) are produced from a common polypeptide precursor (prepro-orexin), through proteolytic processing.

Orexin A is a neuropeptide composed of 33 amino acid with an amino (N)-terminal pyroglutamyl residue, two intra-chain disulphide bonds and carboxy (C)-terminal amidation. Orexin B is a linear neuropeptide sized 28 amino acid, C-terminally amidated. The N-terminal portion has more variability whilst the C-terminal portion is very similar between the two subtypes. The actions of orexins is modulated by their receptors, orexin 1- and orexin 2-receptor (OX1R, OX2R). Orexin 1 receptor has greater affinity for orexin A than B and trasmits signals throughout a G-protein class, which activates a cascade leading to an increase in intracellular calcium. By contrast orexin 2 receptor binds the two subtypes of orexin with similar affinities, and is thought to be associated also to a G inibitory protein class.

These differences indicate that the two types of receptors have different physiological roles[1,18-20]. Furthermore mRNAs receptors show complementary distribution patterns:

- OX1R distributes in prefrontal and infralimbic cortex (IL), hippocampus (CA2), amygdala, bed nucleus of the stria terminalis (BST), PVT, anterior hypothalamus, dorsal raphe (DR), ventral tegmental area (VTA), LC, and laterodorsal tegmental nucleus (LDT)/pedunculopontine nucleus (PPT);

- OX2R distributes in the amygdala, TMN, Arc, dorsomedial hypothalamic nucleus (DMH), paraventricular nucleus (PVN), LHA, BST, PV.T, DR, VTA, LDT/PPT, CA3 in the hippocampus, and medial septal nucleus.

This support the hypothesis that different receptor subtypes play distinct physiological roles. Orexin neurons project to multiple brain region and the wide distribution of its receptors suggests that orexin system is involved in multiple physiological processes such as arousal and sleep, reward, stress, and energy homeostasis.

## Modulation of orexin neurons

Electrophysiological studies on transgenic mice identified several neurotransmitters and neuromodulators influencing the activation or inhibition of orexin neurons activity. For example noradrenaline and serotonin (5HT) inhibit the activity of orexin neurons by sending inhibitory feedback projections. Through its activity on α2-adrenoceptors also dopamine can inhibit orexin neurons, whilst histamine has no effect over them. Furthermore agonists of ionotropic glutamate receptors excite orexin neurons, while glutamate antagonists inhibit their activity, thus indicating that glutamatergic neurons tonically acitvate orexin neurons. Inhibitory stimulation above orexin neurons comes from GABAergic input[21-30]).

Further studies where conducted in order to perform a screening for factors affecting the activity of orexin neurons. Thanks to the help of transgenic mice it has been possible identifying several factors influencing the activity of orexin neurons:

- activators such as cholecystokinin, neurotensin, oxytocin and vasopressin, - inhibitors such as GABA, glucose, 5-HT, noradrenaline, and leptine. Orexine neurons activity should also be modulated by adenosine and concentration in acid and CO2[12, 31-33]. In fact acidification increases neural excitability, whereas alkalinization depresses it.

## Sleep/wake states regulation

Orexin system is important for maintenance of wakefullness, as demonstrated by the fact that narcolepsy is caused by orexin deficiency in human and animals[34-39]. Narcolepsy is a neurological desease affecting ~1 in 2000 individuals in the United State.

Narcolepsy is the results of the loss of orexin-A and orexin-B which have excitatory effects once bound with their receptors. These neuropeptides increase their activity during wakefullness through activation of many aminergic nuclei such as locus coeruleus, raphe nuclei, and tuberomammillary nucleus. The final results of orexins action is a stabilization of wake state and prevention of inappropriate transitions into rapid eye movement (REM) or non-REM sleep, and inhibition of REM sleep[40-48].

In general, orexin sleep/wake regulation may be considered as relevant and essential for cognition because of its specific action in modulation sleep macroarchitecture and NREM sleep instability (Esposito el al., 2013) Moreover, probably due to the same role in sleep modulation, orexin seems to be involved also in migraine pathogenesis.

Moreover, the relationship between good quality of sleep and cognitive performance has been supported by the evidence of a link between the role of non-rapid eye movement sleep and instability in the child's cognitive performance and by mechanisms explaining how learning and cognitive performance depend on a good night's sleep[49-55].

**The role of orexin system among obesity**

Obesity is a clinical condition characterized by a excess of body fat which can lead to negative effects above health such as metabolic syndrome, type 2 diabetes mellitus, coronary heart disease, and finally a reduced life expectancy. In the last decades incidence of obesity has continuing increased in children and adults all over the world causing alert and attention from public health. Environmental and genetic factors cause large variations among humans susceptibility to obesity. Physical activity, and specifically a component named "non-exercise induced thermogenesis" (NEAT), is a factor determining variability. NEAT includes all forms of energy expenditure not associated with formal exercise, such as standing and fidgeting. A complementary concept to that of NEAT is SPA (spontaneous physical activity), used to describe any type of physical activity that does not qualify as voluntary exercise. NEAT and SPA are hereditable not interchangeable, complementary concepts: NEAT refers to energy expenditure while SPA describes the types of physical activity that result in NEAT.

However the regulation of body weight seems to be more complex according to the lack of orexin neurons. Some Authors found body weight loss and protection against obesity in brain mice submitted to repeated orexin A (OXA) injections; OXA has also been discovered to promote SPA and NEAT following injection of orexin into the rostral LH, hypothalamic paraventricular nucleus, nucleus accumbens, locus coeruleus, dorsal raphe nucleus, tuberomammillary nucleus, and substantia nigra[52-60].

**Other function of orexin**

***Feeding Behaviour***

Icv injection of orexins during the light period induces feeding behavior in rodents and zebrafishes, then orexin might regulate feeding behaviour in many species. High concentrations of glucose and leptin hyperpolarize orexin neurons whilst diminuished concentrations of glucose and ghrelin induce depolarization. Lateral hypothalamic area contains neurons which activity is modulated by glucose concentration hence regulating feeding and energy expenditure; then we might hypothesize a predominant role of orexin neurons in these mechanisms. Surprisingly this system permits discrimination of physiological variation in glucose levels due to meals so that orexin neurons might modulate energy balance according to food intake. Human narcolepsy patients shown disregulation in energy homeostasis such as decreased caloric intake and increased BMI thus reflecting feeding abnormalities such as hypophagia and obesity related to impaired thermogenesis[58-66].

**Energy Homeostasis**

Some authors pointed out another regulation of orexin system above muscle glucose metabolism through activation of β2-adrenergic signaling indicaitng a further regulation of orexin above peripheral energy expenditure and not only feeding. A solid state of awake mediated by orexin could also be important in relation to food intake motivation; when facing reduced food availability animals adapt with a longer awake period, revolutionizing their normal pattern of activity. During starvation the activation of orexin neurons mediated by low leptin and glucose levels, might modulate their activity according to energy expenditure and stores, in order to maintain wakefullness, whilst orexin neuron-ablated mice fail to respond to fasting with increased wakefulness and acitvity. This is the confirm that orexin neurons mediate energy balance and arousal, maintaning a consolidated state of wakefulness in hungry animals in order to promote alertness[65-72].

**Reward System**

Recent studies focused their attention on reward system modulation by orexins. Treating narcoleptic patients with amphetamine-like drugs did not lead to addiction to these drugs. Wild-type mice are more susceptible to developing morphine dependence in comparison with orexin knockout mice. Furthermore reward brain circuits in humans affected by narcolepsy were abnormal. In the regulatory mechanisms seem to be clear that orexin neurons modulate reward system and play a predominant role in the mechanism of drug addiction.

Many reports suggest a critical role of orexin signaling in neural plastic effects at glutamatergic synapses in the ventral tegmental area (VTA)[70-75].

## Links with brain

The distribution of orexin neurons in the perifornical area and hypothalamic lateral and posterior area in the rat has been confirmed in humans brain. Orexin-producing neurons are diffusely distributed in multiple brain areas thus modulating the activity of such areas. Orexin neurons projections has been described in the tuberomammillary nucleus (TMN), paraventricular thalamic nucleus (PVT), arcuate nucleus and moreover in the locus ceruleus (LC) as monoaminergic nuclei. It has been confirmed that many orexin-producing neurons are glutamatergic and release glutamate on TMN nucleus. In order to study innervations to orexin neurons, genetically traced mice and rats were used; these studies lead to observe that orexin neurons are linked to several brain areas and nuclei such as lateral parabrachial nucleus (LPB), ventrolateral preoptic nucleus (VLPO), medial and lateral preoptic areas, basal forebrain (BF), posterior/dorsomedial hypothalamus, VTA, DR nucleus, and MnR.

Were furthermore identified associations to areas implied in emotion such as amygdala, nucleus accumbens, and lateral septum; another associtation was found to areas involved in energy homeostasis system originating in the arcuate nucleus. The findings of all these studies confirm that orexin neurons project to brain areas involved in the maintenance of wake/sleep states; orexin neurons are also linked to hypothalamic nuclei regulating feeding and energy homeostasis. This anatomical structure suggests that the activity of orexin neurons influences multiple brain areas[74-93].

In conclusion orexin neurons are multi-tasking neurons and regulate several physiological function such as sleep/wake, feeding, and spontaneous physical activity. SPA and NEAT are key concept in understanding obesity resistance and could become new therapeutic application targets in the near future. Orexin receptors stimulation might be an attractive target of body weight gain correction. Further studies are needed in order to confirm these almost known mechanisms and explore how orexin system regulates emotions, energy homeostasis, feeding behaviour and reward system.

## References


1) Sakurai T, Amemiya A, Ishii M, Matsuzaki I, Chemelli RM, Tanaka H, Williams SC, Richardson JA, Kozlowski GP, Wilson S, Arch JRS, Buckingham RE, Haynes AC, Carr SA, Annan RS, McNulty DE, Liu WS, Terrett JA, Elshourbagy NA, Bergsma DJ, Yanagisawa M (1998) Orexins and orexin receptors: A family of hypothalamic neuropeptides and G protein-coupled receptors that regulate feeding behavior. Cell 92: 573–585. doi: 10.1016/S0092-8674(00)80949-6
2) Chieffi S, Messina G, Villano I, Messina A, Esposito M, Monda V, et al. (2017) Exercise Influence on Hippocampal Function: Possible Involvement of Orexin-A. Front Physiol. 14; 8: 85. doi: 10.3389/fphys.2017.00085.
3) Thannickal TC, Moore RY, Nienhuis R, Ramanathan L, Gulyani S, Aldrich M, Cornford M, Siegel JM (2000) Reduced number of hypocretin neurons in human narcolepsy. Neuron 27: 469-474. doi: 10.1016/S0896-6273(00)00058-1
4) Messina G, De Luca V, Viggiano A, Ascione A, Iannaccone T, et al. (2013) Autonomic nervous system in the control of energy balance and body weight: Personal contributions. Neurol Res Int. 2013: 639280. http://dx.doi.org/10.1155/2013/639280.
5) Messina G, Dalia C, Tafuri D, Monda V, Palmieri F, Dato A, et al. (2014) Orexin-A controls sympathetic activity and eating behavior. Front Psychol. 5: 997. https://doi.org/10.3389/fpsyg.2014.00997.
6) Messina G, Monda V, Moscatelli F, Valenzano AA, Monda G, Esposito T, et al. (2015) Role of Orexin system in obesity. Biol Med (Aligarh) 7: 248. doi: 10.4172/0974-8369.1000248
7) Nambu T, Sakurai T, Mizukami K, Hosoya Y, Yanagisawa M, Goto K (1999) Distribution of orexin neurons in the adult rat brain. Brain Res 827:243–260. doi: 10.1016/S0006-8993(99)01336-0
8) Trivedi P, Yu H, MacNeil DJ, Van der Ploeg LH., Guan X-M (1998) Distribution of orexin receptor mRNA in the rat brain. FEBS Lett 438: 71-75. doi: 10.1016/S0014-5793(98)01266-6
9) Xie X, Crowder TL, Yamanaka A, Morairty SR, Lewinter RD, Sakurai T, Kilduff TS (2006) GABA(B) receptor-mediated modulation of hypocretin/orexin neurones in mouse hypothalamus. J Physiol 574: 399-414. doi: 10.1113/jphysiol.2006.108266
10) Tsunematsu T, Fu L-Y, Yamanaka A, Ichiki K, Tanoue A, Sakurai T, van den Pol AN (2008) Vasopressin increases locomotion through a V1a receptor in orexin/hypocretin neurons: implications for water homeostasis. J Neurosci 28: 228-38. doi: 10.1523/JNEUROSCI.3490-07.2008
11) Liu Z-W, Gao X-B (2007) Adenosine inhibits activity of hypocretin/orexin neurons by the A1 receptor in the lateral hypothalamus: a possible sleep-promoting effect. J Neurophysiol 97: 837-48. doi: 10.1152/jn.00873.2006
12) Williams RH, Jensen LT, Verkhratsky A, Fugger L, Burdakov D (2007) Control of hypothalamic orexin neurons by acid and CO2. Proc Natl Acad Sci U S A 104: 10685-10690. doi: 10.1073/pnas.0702676104
13) Chemelli RM, Willie JT, Sinton CM, Elmquist JK, Scammell T, Lee C, Richardson JA, Clay Williams S,

Xiong Y, Kisanuki Y, Fitch TE, Nakazato M, Hammer RE, Saper CB, Yanagisawa M (1999) Narcolepsy in orexin knockout mice: Molecular genetics of sleep regulation. Cell 98: 437-451. doi: 10.1016/S0092-8674(00)81973-X

14) Mignot E (1998) Genetic and familial aspects of narcolepsy. Neurology 50: S16-22.

15) Saper CB, Chou TC, Scammell TE (2001) The sleep switch: Hypothalamic control of sleep and wakefulness. Trends Neurosci 24:726–731. doi: 10.1016/S0166-2236(00)02002-6

16) Esposito M, Carotenuto M (2013) Intellectual disabilities and power spectra analysis during sleep: a new perspective on borderline intellectual functioning. J Intellect Disabil Res 1-9. doi: 10.1111/jir.12036

17) Rainero I, De Martino P, Pinessi L (2008) Hypocretins and primary headaches: Neurobiology and clinical implications. Expert Rev Neurother 8: 409-416. doi: 10.1586/14737175.8.3.409

18) Perez-Leighton CE, Boland K, Teske J a., Billington C, Kotz CM (2012) Behavioral responses to orexin, orexin receptor gene expression, and spontaneous physical activity contribute to individual sensitivity to obesity. AJP Endocrinol Metab 303: E865-E874. doi: 10.1152/ajpendo.00119.2012

19) Messina G, De Luca V, Viggiano A, Ascione A, Iannaccone T, Chieffi S, Monda M (2013b) Autonomic nervous system in the control of energy balance and body weight: Personal contributions. Neurol Res Int. doi: 10.1155/2013/639280

20) Monda M, Viggiano A, Viggiano A, Mondola R, Viggiano E, Messina G, Tafuri D, De Luca V (2008a) Olanzapine blocks the sympathetic and hyperthermic reactions due to cerebral injection of orexin A. Peptides 29: 120-126. doi: 10.1016/j.peptides.2007.10.016

21) Monda M, Messina G, Mangoni C, De Luca B (2008b) Resting energy expenditure and fat-free mass do not decline during aging in severely obese women. Clin Nutr 27: 657-9. doi: 10.1016/j.clnu.2008.04.005

22) Shiuchi T, Haque MS, Okamoto S, Inoue T, Kageyama H, Lee S, Toda C, Suzuki A, Bachman ES, Kim YB, Sakurai T, Yanagisawa M, Shioda S, Imoto K, Minokoshi Y (2009) Hypothalamic Orexin Stimulates Feeding-Associated Glucose Utilization in Skeletal Muscle via Sympathetic Nervous System. Cell Metab 10: 466-480. doi: 10.1016/j.cmet.2009.09.013

23) Messina G, Palmieri F, Monda V, Messina A, Dalia C, Viggiano A, Tafuri D, Messina A, Moscatelli F, Valenzano A, Cibelli G, Chieffi S, Monda M (2015b) Exercise Causes Muscle GLUT4 Translocation in an Insulin-Independent Manner. Biol Med. doi: http://dx.doi.org/10.4172/0974-8369.1000S3007

24) Di Bernardo G, Messina G, Capasso S, Del Gaudio S, Cipollaro M, Peluso G, Casale F, Monda M, Galderisi U (2014) Sera of overweight people promote in vitro adipocyte differentiation of bone marrow stromal cells. Stem Cell Res Ther 5:4. doi: 10.1186/scrt393

25) Monda M, Messina G, Scognamiglio I, Lombardi A, Martin GA, Sperlongano P, Porcelli M, Caraglia M, Stiuso P (2014) Short-term diet and moderate exercise in young overweight men modulate cardiocyte and hepatocarcinoma survival by oxidative stress. Oxid Med Cell Longev. doi: 10.1155/2014/131024

26) Chieffi S, Iavarone A, Iaccarino L, La Marra M, Messina G, De Luca V, Monda M (2014) Age-related differences in distractor interference on line bisection. Exp brain Res 232: 3659-64. doi: 10.1007/s00221-014-4056-0

27) Viggiano A, Chieffi S, Tafuri D, Messina G, Monda M, De Luca B (2013) Laterality of a second player position affects lateral deviation of basketball shooting. J Sports Sci 37–41. doi: 10.1080/02640414.2013.805236

28) Yang L, Zou B, Xiong X, Pascual C, Xie J, Malik A, et al. (2013) Hypocretin/orexin neurons contribute to hippocampus-dependent social memory and synaptic plasticity in mice. J Neurosc 33: 5275-5284. https://doi.org/10.1523/JNEUROSCI.3200-12.2013.

29) Zhao X, xue Zhang R, Tang S, yan Ren Y, xia Yang W, et al. (2014) Orexin-A-induced ERK ½ activation reverses impaired spatial learning and memory in pentylenetetrazol-kindled rats via OX1R-mediated hippocampal neurogenesis. Peptides 54: 140-147. http://dx.doi.org/10.1016/j.peptides.2013.11.019.

30) Turillazzi E, La Rocca G, Anzalone R, Corrao S, Neri M, Pomara C, et al. Heterozygous nonsense SCN5A mutation W822X explains a simultaneous sudden infant death syndrome. Virchows Arch. 2008; 453(2): 209-16.

31) Turillazzi E, Riezzo I, Neri M, Pomara C, Cecchi R F V. The diagnosis of fatal pulmonary fat embolism using quantitative morphometry and confocal laser scanning microscopy. 2008; 204(4): 259-66.

32) Turillazzi E, Greco P, Neri M, Pomara C, Riezzo I, Fineschi V. Anaphylactic latex reaction during anaesthesia: The silent culprit in a fatal case. Forensic Sci Int. 2008; 179(1).

33) Pomara C, Fiore C, D'Errico S, Riezzo I, Fineschi V. Calcium oxalate crystals in acute ethylene glycol poisoning: A confocal laser scanning microscope study in a fatal case. Clin Toxicol. 2008; 46(4): 322-4.

34) Cerretani D, Bello S, Cantatore S, Fiaschi AI, Montefrancesco G, Neri M, et al. Acute administration of 3,4-methylenedioxymethamphetamine (MDMA) induces oxidative stress, lipoperoxidation and TNFα-mediated apoptosis in rat liver. Pharmacol Res. 2011; 64(5): 517-27.

35 Pomara C, Cassano T, D'Errico S, Bello S, Romano AD, Riezzo I, et al. Data Available on the Extent of Cocaine Use and Dependence: Biochemistry, Pharmacologic Effects and Global Burden of Disease of Cocaine Abusers. Curr Med Chem. 2012; 19(33): 5647-57.

36) Sperandeo R, Maldonato M, Baldo G, Dell'Orco S. Executive functions, temperament and character traits: A quantitative analysis of the relationship between personality and prefrontal functions. In: 7th IEEE International Conference on Cognitive Infocommunications, CogInfoCom 2016 - Proceedings. 2017. p. 43-–8.

37) Turillazzi E, Di Peri GP, Nieddu A, Bello S, Monaci F, Neri M, et al. Analytical and quantitative concentration of gunshot residues (Pb, Sb, Ba) to estimate entrance hole and shooting-distance using confocal laser microscopy and inductively coupled plasma atomic emission spectrometer analysis: An experimental study. Forensic Sci Int. 2013; 231(1-3):142-9.

38) Pomara C, Barone R, Marino Gammazza A, Sangiorgi C, Barone F, Pitruzzella A, et al. Effects of Nandrolone

Stimulation on Testosterone Biosynthesis in Leydig Cells. J Cell Physiol. 2016; 231(6): 1385-91.

39) Pomara C, Gianpaolo DP, Monica S, Maglietta F, Sessa F, Guglielmi G, et al. "Lupara Bianca" a way to hide cadavers after Mafia homicides. A cemetery of Italian Mafia. A case study. Leg Med. 2015; 17(3): 192–7.

40) Maldonato M. The archipelago of consciousness. Bright Sussex Acad Press. 2015;

41. Viggiano A, Nicodemo U, Viggiano E, Messina G, Viggiano A, Monda M, et al. Mastication overload causes an increase in O 2 - production into the subnucleus oralis of the spinal trigeminal nucleus. Neuroscience. 2010; 166(2): 416-21.

42) Messina G, Viggiano A, Tafuri D, Palmieri F, De Blasio S, Messina A, et al. Role of orexin in obese patients in the intensive care unit. J Anesth Clin Res. 2014; 5(3).

43) Ruberto, M., Precenzano, F., Parisi, L., Salerno, M., Agata M, Messina, G. Roccella M. Visuomotor integration skills in children affected by obstructive sleep apnea syndrome: A case-control studyActa. Medica Mediterr. 2016; 32(5): 1659-63.

44) F Precenzano, M Ruberto, L Parisi, M Salerno, A Maltese ID. ADHD-like symptoms in children affected by obstructive sleep apnea syndrome: a case-control study. Acta Medica Mediterr. 2016; 32: 1755-9.

45) F Precenzano, M Ruberto, L Parisi, M Salerno, A Maltese, C Vagliano. Executive functioning in preschool children affected by autism spectrum disorder: a pilot study. Acta Medica Mediterr. 2017; 33-5.

46) Messina G, Zannella C, Monda V, Dato A, Liccardo D, S DB, et al. The Beneficial Effects of Coffee in Human Nutrition. 2015; 7(4).

47) Francesco Precenzano, Paola Lombardi, Maria Ruberto, Lucia Parisi, Margherita salerno, A. Maltese, Ilaria D'Alessandro, Immacolata Della Valle, Rosaria Martina Magliulo GM, Roccella M. Internalizing Symptoms in children affecyed by childhood absence apilepsy: a preliminar study. Acta Medica Mediterr. 2016; 32: 750-3.

48) Carotenuto M, Esposito M, Precenzano F, Castaldo L, Roccella M. Cosleeping in childhood migraine. Minerva Pediatr. 2011; 63(2): 105-9.

49) Messina A, De Fusco C, Monda V, Esposito M, Moscatelli F, Valenzano A, et al. Role of the Orexin System on the Hypothalamus-Pituitary-Thyroid Axis. Front Neural Circuits. 2016; 10: 66.

50) Messina G, Di Bernardo G, Viggiano A, De Luca V, Monda V, Messina A, et al. Exercise increases the level of plasma orexin A in humans. J Basic Clin Physiol Pharmacol. 2016 Nov 1; 27(6): 611-6.

51) Carotenuto M, Messina A, Monda V, Precenzano F, Iacono D, Verrotti A, et al. Maternal Stress and Coping Strategies in Developmental Dyslexia: An Italian Multicenter Study. Front Psychiatry. 2017 Dec 22;8.

52) Neri V, Ambrosi A, Fersini A, Tartaglia N, Lapolla F, Forlano I. Severe acute pancreatitis: clinical forms of different gravity. Ann Ital Chir. 2013; 84(1): 47-53.

53) Neri V, Fersini A, Ambrosi A, Tartaglia N, Valentino TP. Diagnostic evaluation prior to cholecystectomy in mild-moderate acute biliary pancreatitis. Ann Ital Chir [Internet]. 80(5): 363-7. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20131548

54) Cianci P, Tartaglia N, Altamura A, Fersini A, Vovola F, Sanguedolce F, et al. A recurrent epidermoid cyst of the spleen: Report of a case and literature review. World J Surg Oncol. 2016; 14(1).

55) Esposito M, Messina A, Monda V, Bitetti I, Salerno F, Precenzano F, Pisano S, Salvati T, Gritti A, Marotta R, Lavano SM, Lavano F, Maltese A, Parisi L, Salerno M, Tripi G, Gallai B, Roccella M, Bove D, Ruberto M, Toraldo R MG and CM. The Rorschach Test Evaluation in Chronic Childhood Migraine: A Preliminary Multicenter Case-Control Study. Front Neurol. 2017; 8(680).

56) Forlano I, Fersini A, Tartaglia N, Ambrosi A, Neri V. Biliary papillomatosis. Case report. Ann Ital Chir. 82(5): 405-8.

57) Neri V, Fersini A, Ambrosi A, Tartaglia N, Valentino TP. Mild-moderate acute biliary pancreatitis: Role of magnetic resonance cholangiopancreatography in preparation of cholecystectomy. Vol. 38, Pancreas. 2009. p. 717.

58) Salomone F, Li Volti G, Vitaglione P, Morisco F, Fogliano V, Zappalà A, et al. Coffee enhances the expression of chaperones and antioxidant proteins in rats with nonalcoholic fatty liver disease. Transl Res. 2014; 163(6): 593-602.

59) Esposito M, Gallai B, Parisi L, Castaldo L, Marotta R, Lavano SM, et al. Self-concept evaluation and migraine without aura in childhood. Neuropsychiatr Dis Treat. 2013; 9: 1061-6.

60) Bramanti V, Tomassoni D, Grasso S, Bronzi D, Napoli M, Campisi A, et al. Cholinergic precursors modulate the expression of heme oxigenase-1, p21 during astroglial cell proliferation and differentiation in culture. Neurochem Res. 2012; 37(12): 2795-804.

61) Avola R, Di Tullio MA, Fisichella A, Tayebati SK, Tomassoni D. Glial fibrillary acidic protein and vimentin expression is regulated by glucocorticoids and neurotrophic factors in primary rat astroglial cultures. In: Clinical and Experimental Hypertension. 2004. p. 323-33.

62) Turillazzi E, Vacchiano G, Luna-Maldonado A, Neri M, Pomara C, Rabozzi R, et al. Tryptase, CD15 and IL-15 as reliable markers for the determination of soft and hard ligature marks vitality. Histol Histopathol. 2010; 25(12): 1539-46.

63) Vyshka G, Vacchiano G. Severe flaccid paraparesis following spinal anaesthesia: A sine materia occurrence. BMJ Case Rep. 2014;

64) Vacchiano G, Vyshka G. Balkans syndrome: A potential link between multiple sclerosis and hypervaccination. J R Army Med Corps. 2012; 158(4): 338-40.

65) Esposito M, Marotta R, Gallai B, Parisi L, Patriciello G, Lavano SM, et al. Temperamental characteristics in childhood migraine without aura:A multicenter study. Neuropsychiatr Dis Treat. 2013; 9: 1187-92.

66. Esposito M, Carotenuto M. Ginkgolide B complex efficacy for brief prophylaxis of migraine in school-aged children: An open-label study. Neurol Sci. 2011; 32(1): 79-81.

67) Esposito M, Pascotto A, Gallai B, Parisi L, Roccella M, Marotta R, et al. Can headache impair intellectual abilities in children? an observational study. Neuropsychiatr Dis Treat. 2012; 8: 509-13.

68) Carotenuto M, Esposito M. Nutraceuticals safety and efficacy in migraine without aura in a population of children affected by neurofibromatosis type I. Neurol

Sci. 2013; 34(11): 1905-9.
69) Monda V, La Marra M, Perrella R, Caviglia G, Iavarone A, Chieffi S, Messina G, Carotenuto M, Monda M, Messina A.Obesity and brain illness: from cognitive and psychological evidences to obesity paradox. Diabetes Metab Syndr Obes. 2017 Nov 21; 10: 473-479. doi: 10.2147/DMSO.S148392.
70) Chieffi S, Messina G, Messina A, Villano I, Monda V, Ambra FI, Garofalo E, Romano F, Mollica MP, Monda M, Iavarone A.Memory for Spatial Locations in a Patient with Near Space Neglect and Optic Ataxia: Involvement of the Occipitotemporal Stream. Front Neurol. 2017 May 31; 8: 231. doi: 10.3389/fneur.2017.00231.
71) Chieffi S, Carotenuto M, Monda V, Valenzano A, Villano I, Precenzano F, Tafuri D, Salerno M, Filippi N, Nuccio F, Ruberto M, De Luca V, Cipolloni L, Cibelli G, Mollica MP, Iacono D, Nigro E, Monda M, Messina G, Messina A.Orexin System: The Key for a Healthy Life. Front Physiol. 2017 May 31; 8: 357. doi: 10.3389/fphys.2017.00357.
72) Villano I, Messina A, Valenzano A, Moscatelli F, Esposito T, Monda V, Esposito M, Precenzano F, Carotenuto M, Viggiano A, Chieffi S, Cibelli G, Monda M, Messina G. Basal Forebrain Cholinergic System and Orexin Neurons: Effects on Attention. Front Behav Neurosci. 2017 Jan 31; 11: 10. doi: 10.3389/fnbeh.2017.00010.
73) Fiorelli A, Messina G, Chiodini P, Costanzo S, Viggiano A, Monda M, Vicidomini G, Santini M. Cardiac Autonomic Changes After Thoracic Sympathectomy: A Prospective, Randomized Study. Ann Thorac Surg. 2017 Jan; 103(1): 216-224. doi: 10.1016/j.athoracsur.2016.10.055.
74) Esposito T, Lobaccaro JM, Esposito MG, Monda V, Messina A, Paolisso G, Varriale B, Monda M, Messina G. Effects of low-carbohydrate diet therapy in overweight subjects with autoimmune thyroiditis: possible synergism with ChREBP. Drug Des Devel Ther. 2016 Sep 14; 10: 2939-2946.
75) Messina A, De Fusco C, Monda V, Esposito M, Moscatelli F, Valenzano A, Carotenuto M, Viggiano E, Chieffi S, De Luca V, Cibelli G, Monda M, Messina G. Role of the Orexin System on the Hypothalamus-Pituitary-Thyroid Axis. Front Neural Circuits. 2016 Aug 25; 10: 66. doi: 10.3389/fncir.2016.00066.
76) Messina G, Chieffi S, Viggiano A, Tafuri D, Cibelli G, Valenzano A, Triggiani AI, Messina A, De Luca V, Monda M. Parachute Jumping Induces More Sympathetic Activation Than Cortisol Secretion in First-Time Parachutists. Asian J Sports Med. 2016 Mar 8; 7(1): e26841. doi: 10.5812/asjsm.26841.
77) Messina G, De Luca V, Viggiano A, Ascione A, Iannaccone T, Chieffi S, Monda M. Autonomic nervous system in the control of energy balance and body weight: personal contributions. Neurol Res Int. 2013; 2013: 639280. doi: 10.1155/2013/639280.
78) Sperandeo R, Monda V, Messina G, Carotenuto M, Maldonato NM, Moretto E, Leone E, De Luca V, Monda M, Messina A . Brain functional integration: an epidemiologic study on stress-producing dissociative phenomena. Neuropsychiatric Disease and Treatment 2018: 14 11-19
79) Messina G, Vicidomini C, Viggiano A, Tafuri D, Cozza V, Cibelli G, Devastato A, De Luca B, Monda M. Enhanced parasympathetic activity of sportive women is paradoxically associated to enhanced resting energy expenditure. Auton Neurosci. 2012 Aug 16; 169(2): 102-6. doi: 10.1016/j.autneu.2012.05.003.
80) Viggiano A, Vicidomini C, Monda M, Carleo D, Carleo R, Messina G, Viggiano A, Viggiano E, De Luca B. Fast and low-cost analysis of heart rate variability reveals vegetative alterations in noncomplicated diabetic patients. J Diabetes Complications. 2009 Mar-Apr; 23(2): 119-23. doi: 10.1016/j.jdiacomp.2007.11.009.
81) Carotenuto M, Gimigliano F, Fiordelisi G, Ruberto M, Esposito M. Positional abnormalities during sleep in children affected by obstructive sleep apnea syndrome: The putative role of kinetic muscular chains. Med Hypotheses. 2013; 81(2): 306-8.
82) Esposito M, Marotta R, Gallai B, Parisi L, Patriciello G, Lavano SM, Mazzotta G,
Roccella M, Carotenuto M. Temperamental characteristics in childhood migraine without aura: a multicenter study. Neuropsychiatr Dis Treat. 2013; 9: 1187-92. doi: 10.2147/NDT.S50458
83) Esposito M, Gallai B, Parisi L, Castaldo L, Marotta R, Lavano SM, Mazzotta G, Roccella M, Carotenuto M. Self-concept evaluation and migraine without aura in childhood. Neuropsychiatr Dis Treat. 2013; 9: 1061-6. doi: 10.2147/NDT.S49364
84) Carotenuto M, Esposito M, Di Pasquale F, De Stefano S, Santamaria F. Psychological, cognitive and maternal stress assessment in children with primary ciliary dyskinesia. World J Pediatr. 2013 Nov; 9(4): 312-7. doi: 10.1007/s12519-013-0441-1
85) Perillo L, Esposito M, Contiello M, Lucchese A, Santini AC, Carotenuto M. Occlusal traits in developmental dyslexia: a preliminary study. Neuropsychiatr Dis Treat. 2013;9:1231-7. doi: 10.2147/NDT.S49985.
86) Moscatelli F, Valenzano A, Petito A, Triggiani AI, Ciliberti MAP, Luongo L, Carotenuto M, Esposito M, Messina A, Monda V, Monda M, Capranica L, Messina G, Cibelli G. Relationship between blood lactate and cortical excitability between taekwondo athletes and non-athletes after hand-grip exercise. Somatosens Mot Res. 2016 Jun; 33(2): 137-44. doi: 10.1080/08990220.2016.1203305
87) Verrotti A, Carotenuto M, Altieri L, Parisi P, Tozzi E, Belcastro V, Esposito M, Guastaferro N, Ciuti A, Mohn A, Chiarelli F, Agostinelli S. Migraine and obesity: metabolic parameters and response to a weight loss programme. Pediatr Obes. 2015 Jun; 10(3): 220-5. doi: 10.1111/ijpo.245
88) Pasquali D, Carotenuto M, Leporati P, Esposito M, Antinolfi L, Esposito D, Accardo G, Carella C, Chiovato L, Rotondi M. Maternal hypothyroidism and subsequent neuropsychological outcome of the progeny: a family portrait. Endocrine. 2015 Dec;50(3):797-801. doi: 10.1007/s12020-015-0564-3
89) Esposito M, Ruberto M, Gimigliano F, Marotta R, Gallai B, Parisi L, Lavano SM, Roccella M, Carotenuto M. Effectiveness and safety of Nintendo Wii Fit Plus™ training in children with migraine without aura: a preliminary study. Neuropsychiatr Dis Treat. 2013; 9: 1803-10. doi: 10.2147/NDT.S53853
90) Capovilla G, Beccaria F, Montagnini A, Cusmai R, Franzoni E, Moscano F, Coppola G, Carotenuto M,

Gobbi G, Seri S, Nabbout R, Vigevano F. Short-term nonhormonal and nonsteroid treatment in West syndrome. Epilepsia. 2003 Aug;44(8): 1085-8. Erratum in: Epilepsia. 2004 Jul; 45(7):887

91) Esposito M, Precenzano F, Sorrentino M, Avolio D, Carotenuto M. A Medical Food Formulation of Griffonia simplicifolia/Magnesium for Childhood Periodic Syndrome Therapy: An Open-Label Study on Motion Sickness. J Med Food. 2015 Aug; 18(8): 916-20. doi: 10.1089/jmf.2014.0113

92) Elia M, Amato C, Bottitta M, Grillo L, Calabrese G, Esposito M, Carotenuto M. An atypical patient with Cowden syndrome and PTEN gene mutation presenting with cortical malformation and focal epilepsy. Brain Dev. 2012 Nov; 34(10): 873-6. doi: 10.1016/j.braindev.2012.03.005

93) Morandi A, Bonnefond A, Lobbens S, Carotenuto M, Del Giudice EM, Froguel P, Maffeis C. A girl with incomplete Prader-Willi syndrome and negative MS-PCR, found to have mosaic maternal UPD-15 at SNP array. Am J Med Genet A. 2015 Nov; 167A(11): 2720-6. doi: 10.1002/ajmg.a.37222

*Corresponding Author:*
Dr. ANNA VALENZANO
e-mail: anna.valenzano@unifg.it
Department of Clinical and Experimental Medicine, University of Foggia
Foggia
*(Italy)*