



<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

Melatonin therapy in Duchenne's muscular dystrophy



Darío Acuña Castroviejo

**RESEARCH GROUP:
CTS-101: INTERCELLULAR COMMUNICATION**

8th International Annual Duchenne Conference
London, November 12-13 2010



<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

DMD:

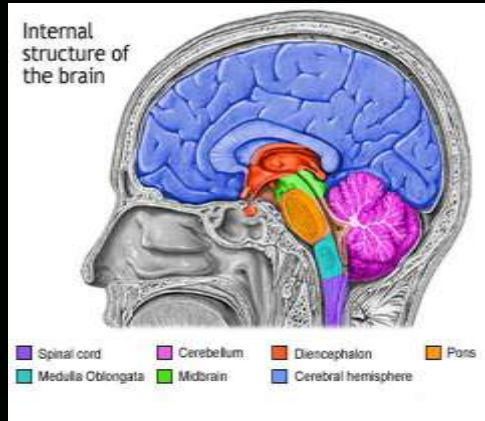
CIRCADIAN RHYTHMS DISORDERS

OXIDATIVE STRESS AND INFLAMMATION

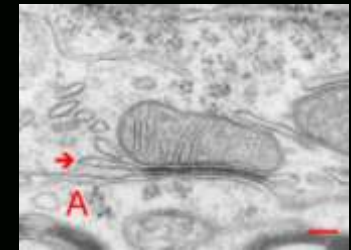


<http://melatonin.ugr.es>
dacuna@ugr.es

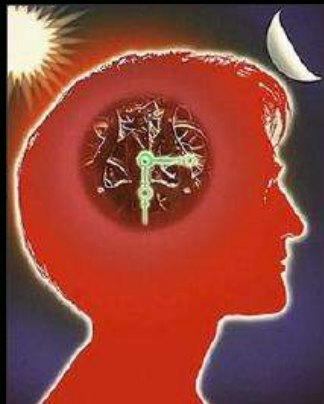
INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN



40% REDUCTION IN NUMBER
REMAINDER UNCOUPLED



BLUNTS CIRCADIAN RHYTHMS:
Melatonin
sleep/wake



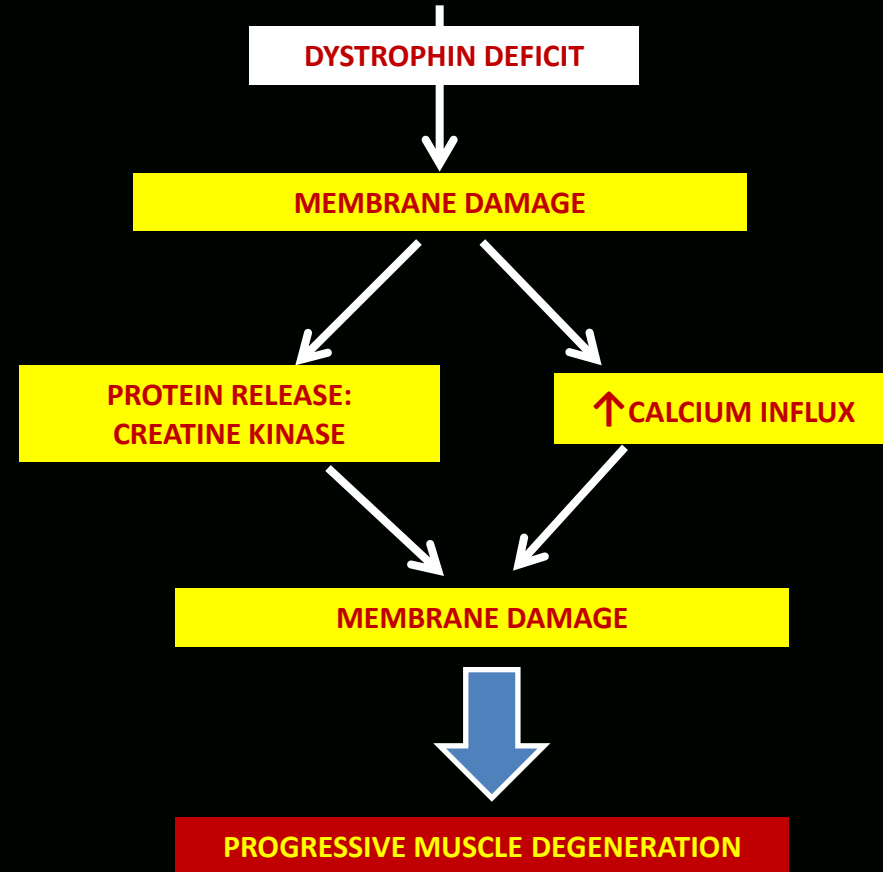
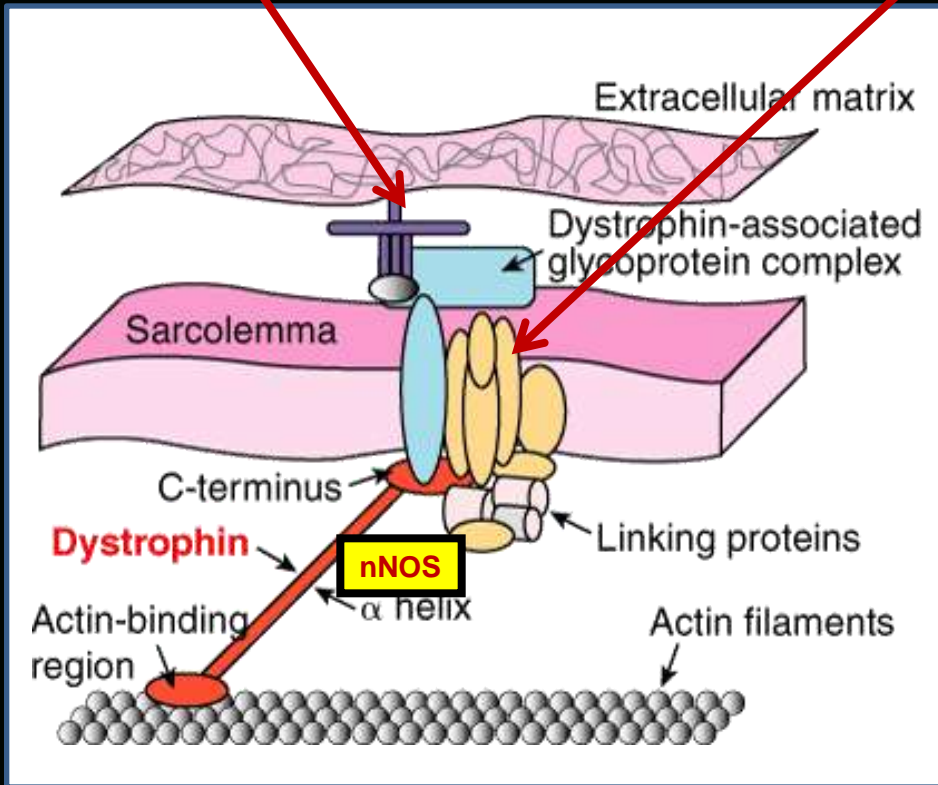
CLOCK and BMAL1

DMD



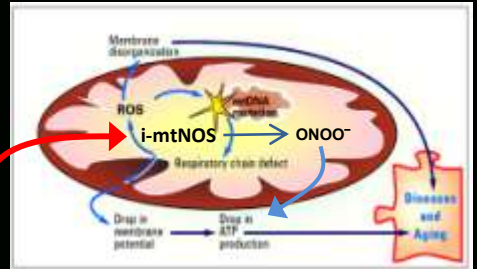
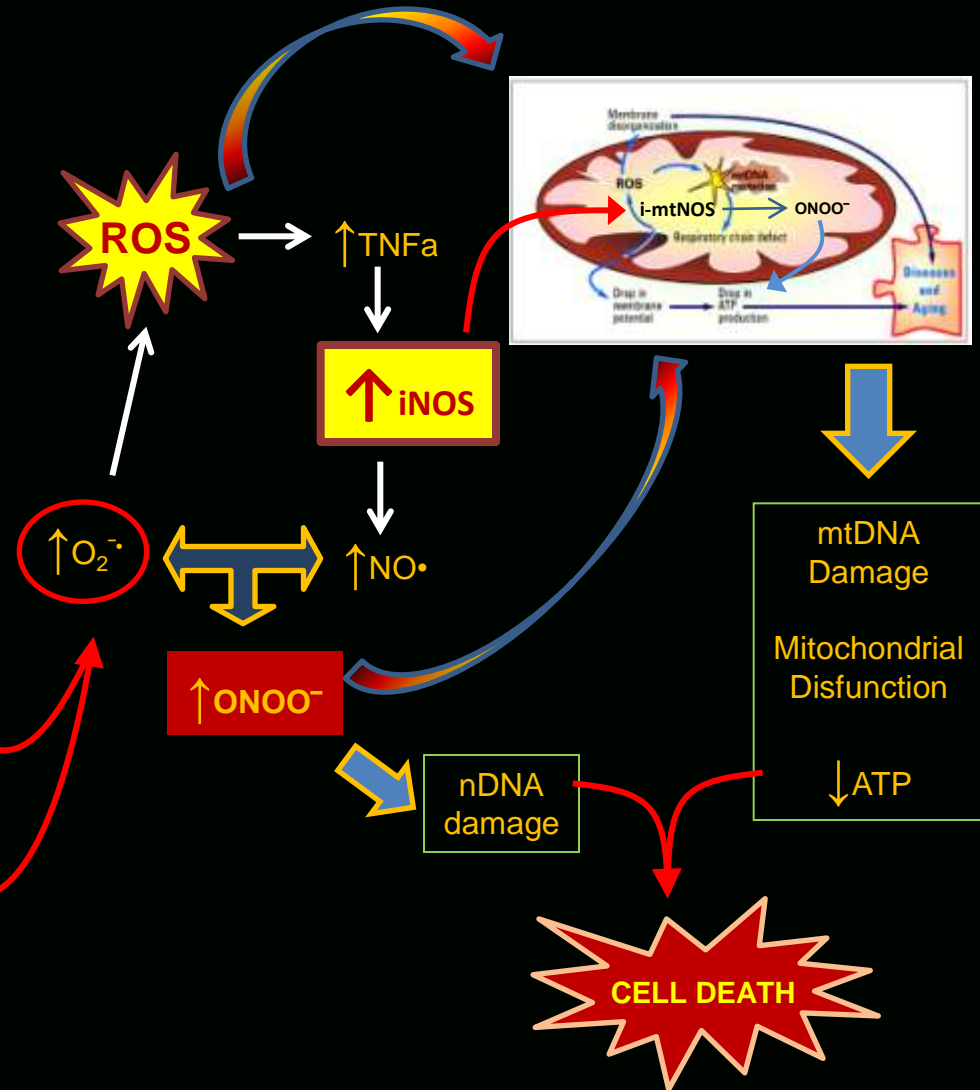
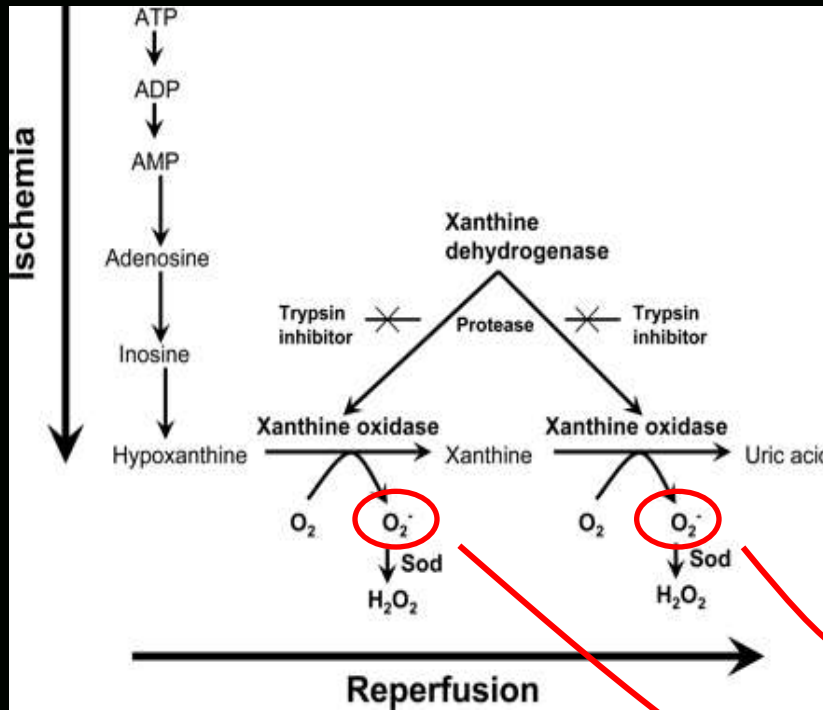
Laminin $\alpha 2$ (merosin)

**DYSTROPHIN-GLYCOPROTEIN COMPLEX (DGC):
PREVENT MUSCLE CONTRACTION-RELAXATION DAMAGE**





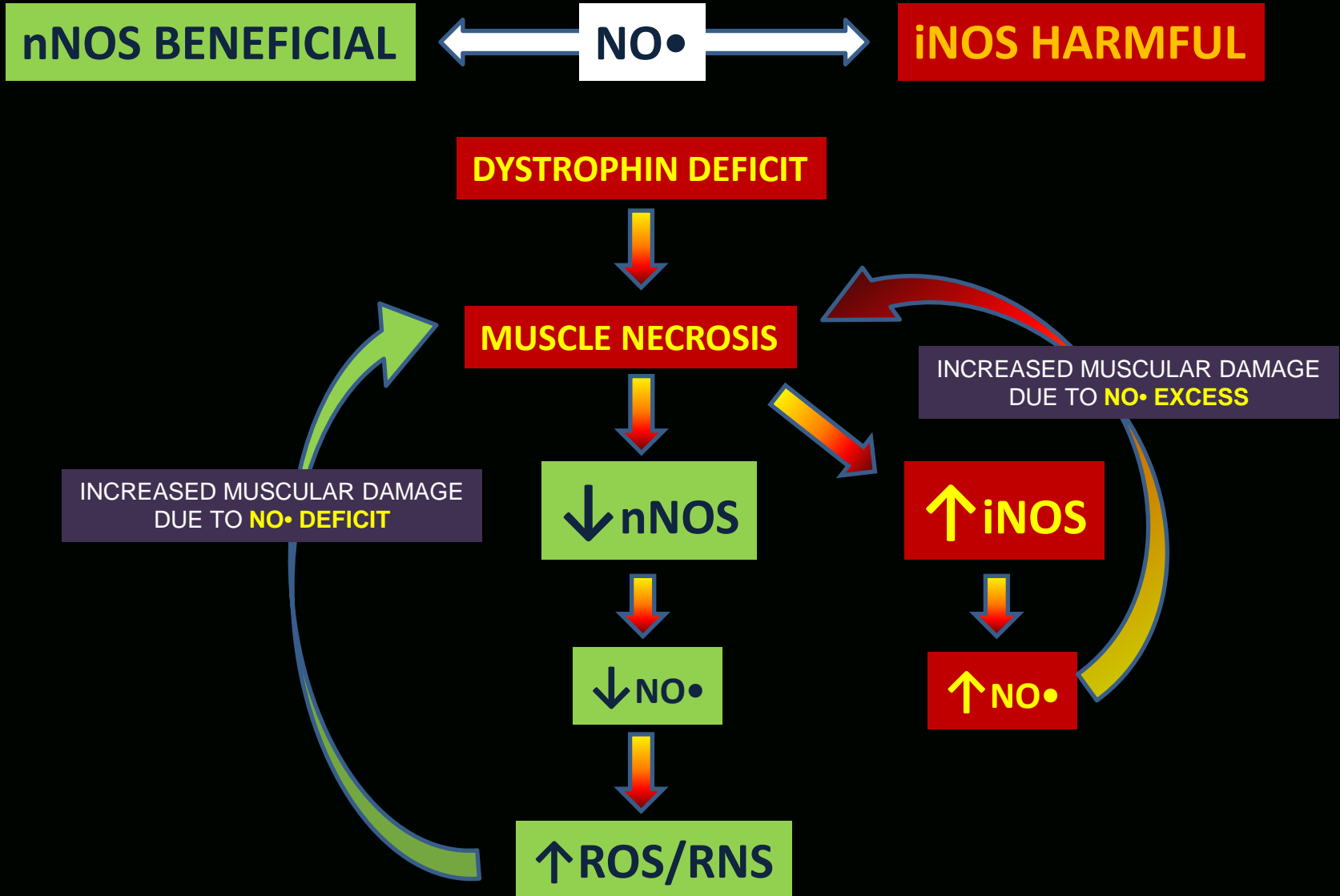
REPERFUSION-INDUCED ROS/RNS





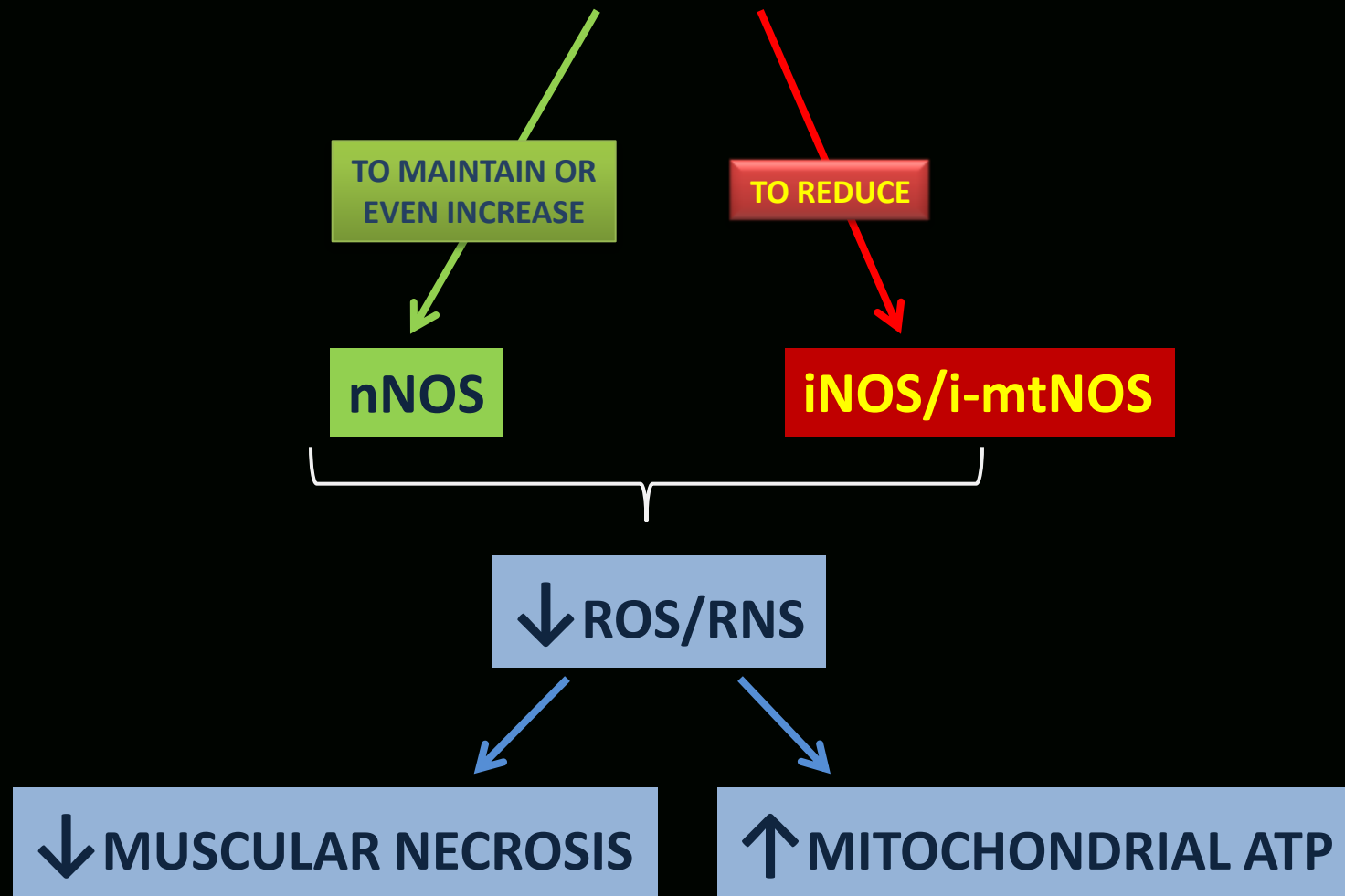
<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN





THUS, TO REDUCE MUSCULAR NECROSIS IN DMD



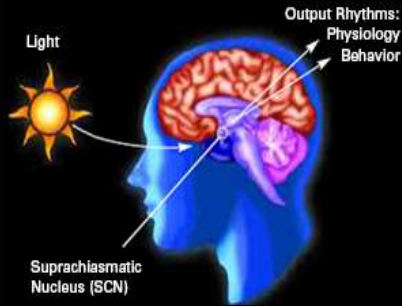


<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

MELATONIN: AN ANTIOXIDANT AND ANTIINFLAMMATORY MOLECULE





SOURCES OF MELATONIN

Pineal aMT:

$[aMT]_{\text{serum}} < 0.5 \text{ nM}$

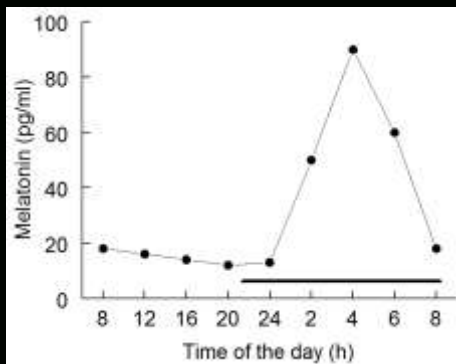
- Endogenous synchronizer
- Crosses all cellular barriers

Extrapineal aMT:

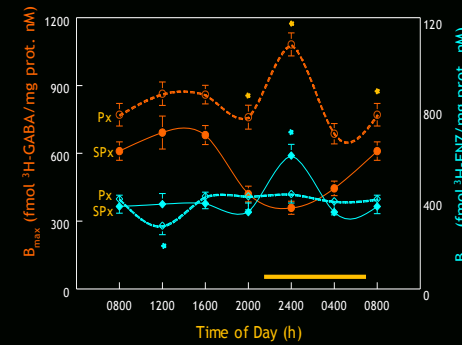
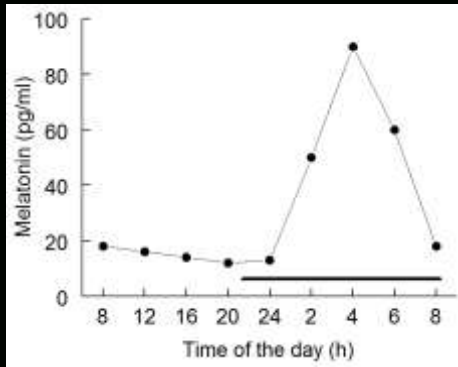
$[aMT]_{\text{tissue}} < 1-10 \mu\text{M}$

- Harderian gland
- Retina
- Immune system
- Intestinal epithelium
- Ovary
- Testicles
- Bone marrow
- Cerebrospinal fluid
- Bile
- NAT and HIOMT are expressed in all tissues

?



**ANTIOXIDANT
ANTI-INFLAMMATORY**



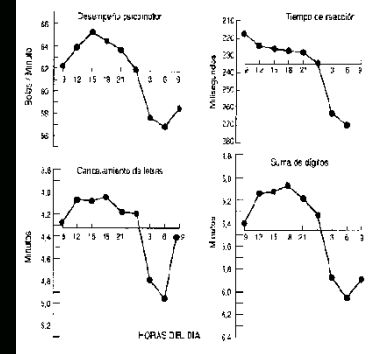
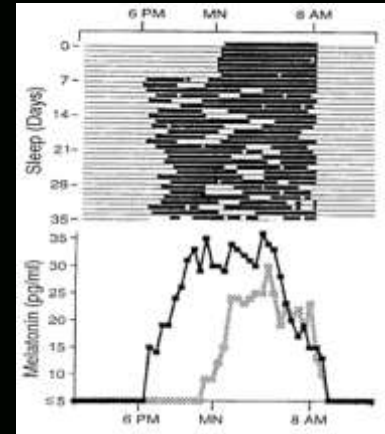
ENDOCRINE RHYTHMS:

- ACTH/cortisol*
- Thyroid hormones*
- GH*

NON-ENDOCRINE RHYTHMS:

- Neurotransmitters*
- Sleep/wake*
- Activity/rest*

CLOCK
and
BMAL1

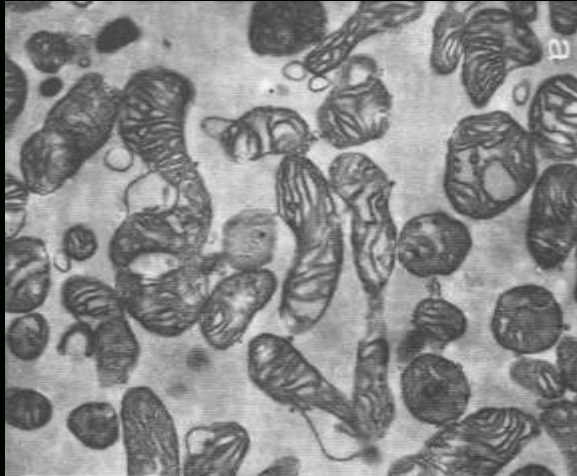




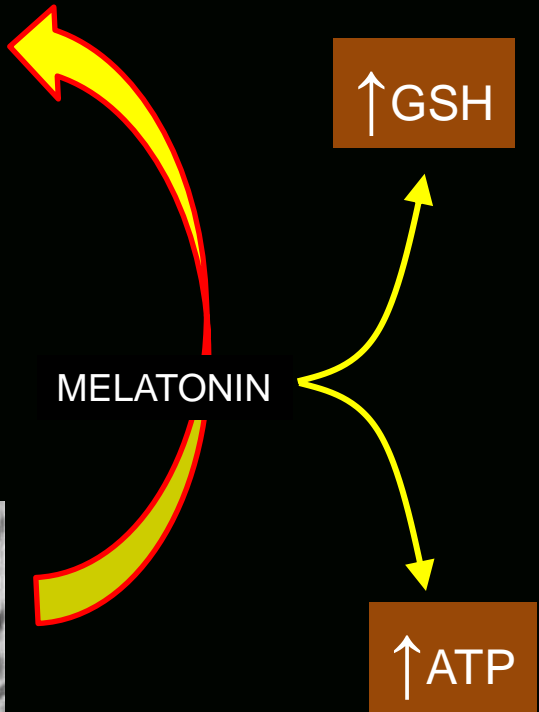
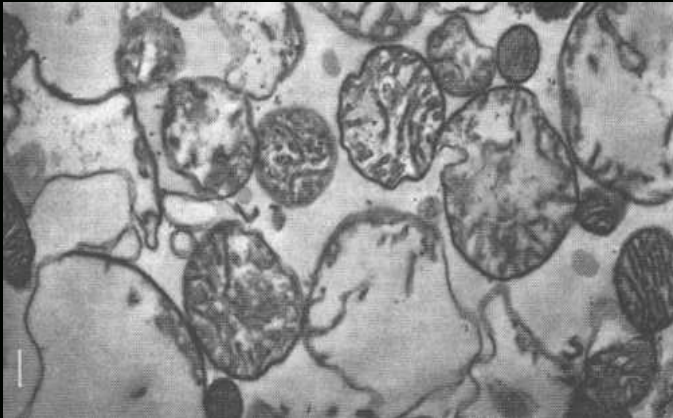
<http://melatonin.ugr.es>
 dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
 BIOMEDICAL RESEARCH CENTER
 HEALTH SCIENCE TECHNOLOGY PARK
 UNIVERSITY OF GRANADA, SPAIN

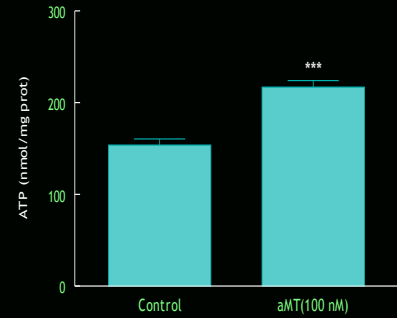
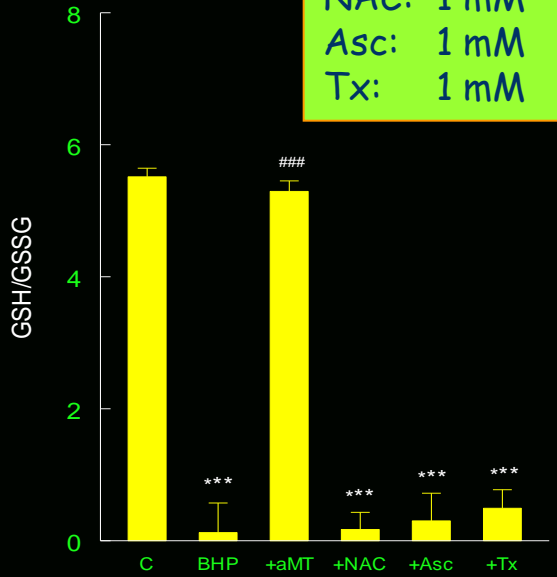
MELATONIN: ANTIOXIDANT



BHP



aMT: 100 nM
 NAC: 1 mM
 Asc: 1 mM
 Tx: 1 mM



Martín et al., *FASEB J* 14:1677-1679, 2000
 Martín et al., *Int J Biochem Cell Biol* 34:348-357, 2002



<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

MELATONIN: ANTI-INFLAMMATORY

FASEB J,13:1537-1546, 1999

Melatonin inhibits expression of the inducible NO synthase II in liver and lung and prevents endotoxemia in lipopolysaccharide-induced multiple organ dysfunction syndrome in rats

ELENA CRESPO,* MANUEL MACÍAS,* DAVID POZO,† GERMAINE ESCAMES,* MIGUEL MARTÍN,* FRANCISCO VIVES,* JUAN M. GUERRERO,† AND DARIO ACUÑA-CASTROVIEJO*¹

*Departamento de Fisiología, Instituto de Biotecnología, Universidad de Granada, Spain;

†Departamento de Bioquímica Médica y Biología Molecular, Facultad de Medicina, Universidad de Sevilla, Spain

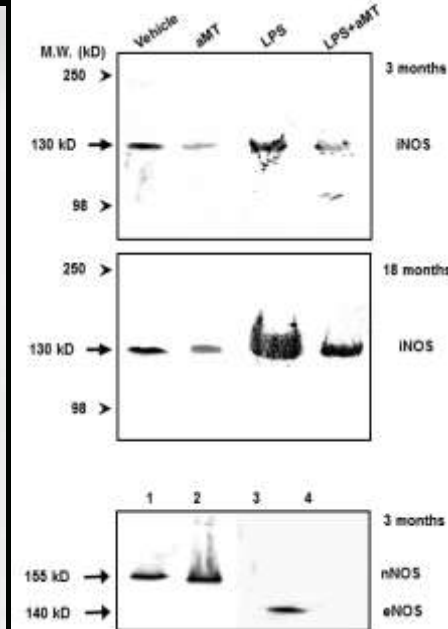
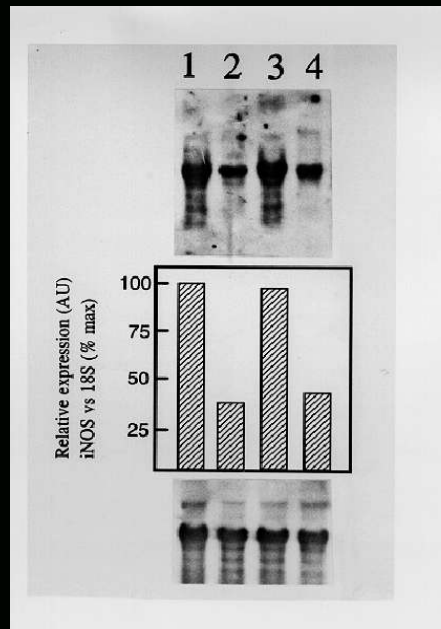
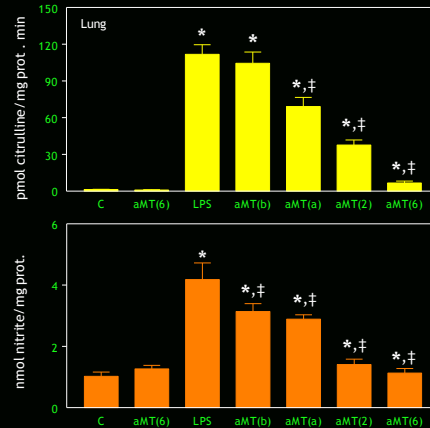
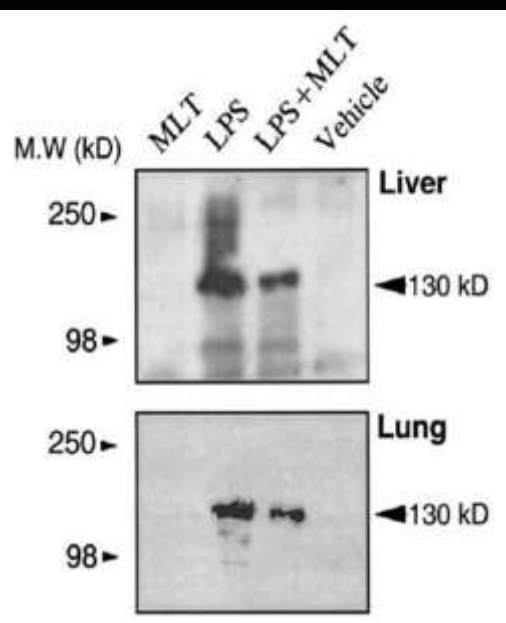
The FASEB Journal express article 10.1096/fj.0692fje. Published online March 28, 2003.

Melatonin counteracts lipopolysaccharide-induced expression and activity of mitochondrial nitric oxide synthase in rats

Germaine Escames, Josefa León, Manuel Macías, Huda Khaldy, and Dario Acuña-Castroviejo

Departamento de Fisiología, Instituto de Biotecnología, Universidad de Granada, Granada, Spain

Corresponding author: Dario Acuña-Castroviejo, Departamento de Fisiología, Facultad de Medicina, Avda. de Madrid 11, E-18012 Granada, Spain. E-mail: dacuna@ugr.es





<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

MELATONIN THERAPY

IN DMD



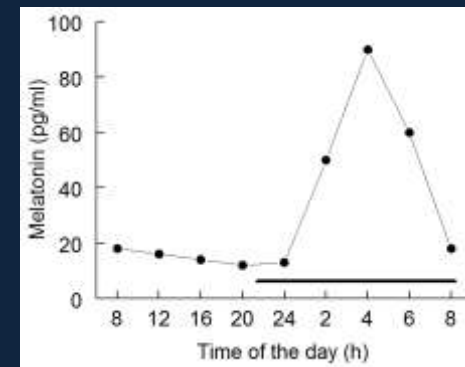
MELATONIN IN DMD

-10 patients 12.8 ± 0.98 years old, Neuropediatric Unit, University Hospital of Granada, Spain

-Genetic diagnoses of DMD

-MELATONIN:

-60 mg at night + 10 mg at morning



BLOOD SAMPLES:

-Antecubital vein at 9:00 hr before, and 3, 6, and 9 mo. after melatonin

-Plasma and erythrocyte aliquots were frozen at -80°C



<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

OXIDATIVE/NITROSATIVE STRESS MARKERS

PLASMA:

EXTRACELLULAR MARKERS

CELL MEMBRANE DAMAGE: **LIPID PROXIDATION (LPO)**

INFLAMMATORY RESPONSE: **NITRITES (NO_x) and CYTOKINES (IL-1 β , IL-2, IL-6, INF γ , TNF α)**

ERYTHROCYTES:

INTRACELLULAR MARKERS

FREE RADICAL PREVENTION: **SUPEROXIDE DISMUTASE (SOD)**

ANTIOXIDATIVE DEFENSE: **REDUCED (GSH) AND OXIDIZED (GSSG) GLUTATHIONE**

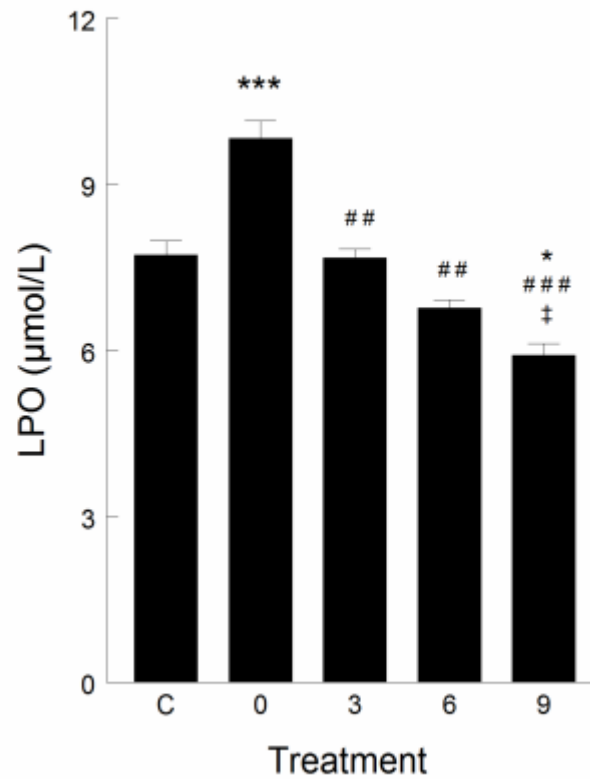
GSH REDOX CYCLE: **GLUTATHIONE PEROXIDASE (GP_x) AND REDUCTASE (GR_d)**



<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

OXIDATIVE STRESS MARKERS

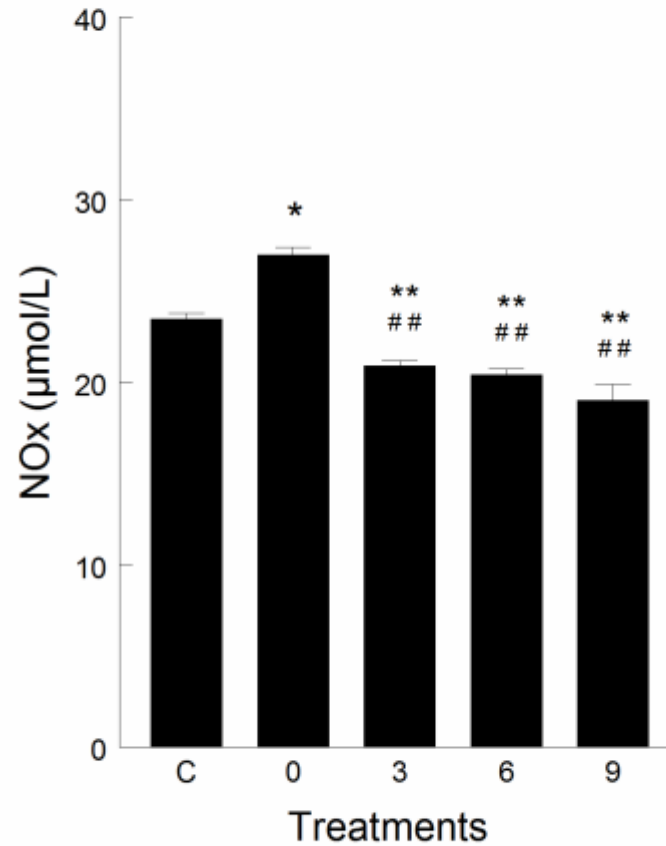




<http://melatonin.ugr.es>
dacuna@ugr.es

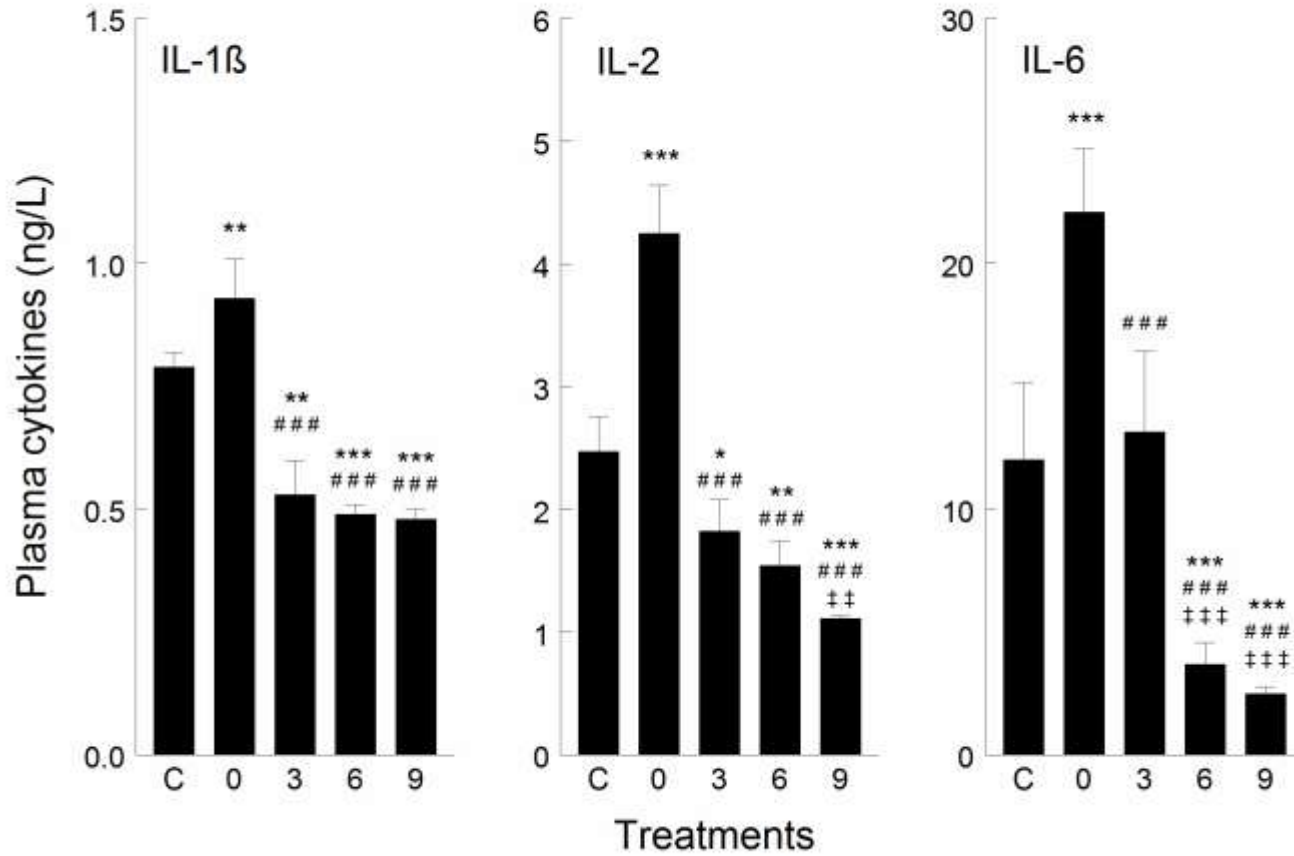
INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

INFLAMMATION MARKERS





INFLAMMATION MARKERS





INFLAMMATION MARKERS

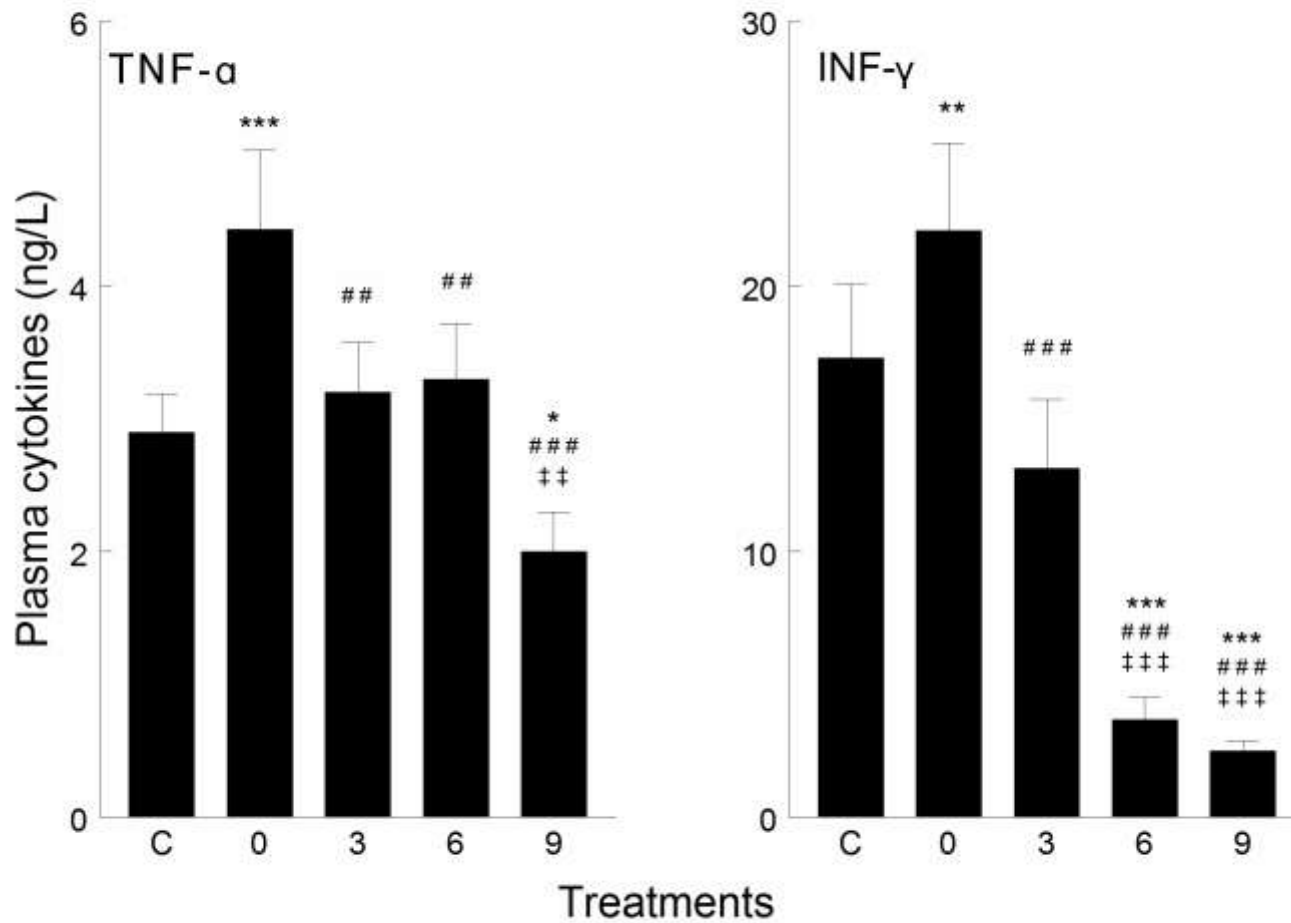




Table 1. Biochemical data obtained from DMD patients and controls

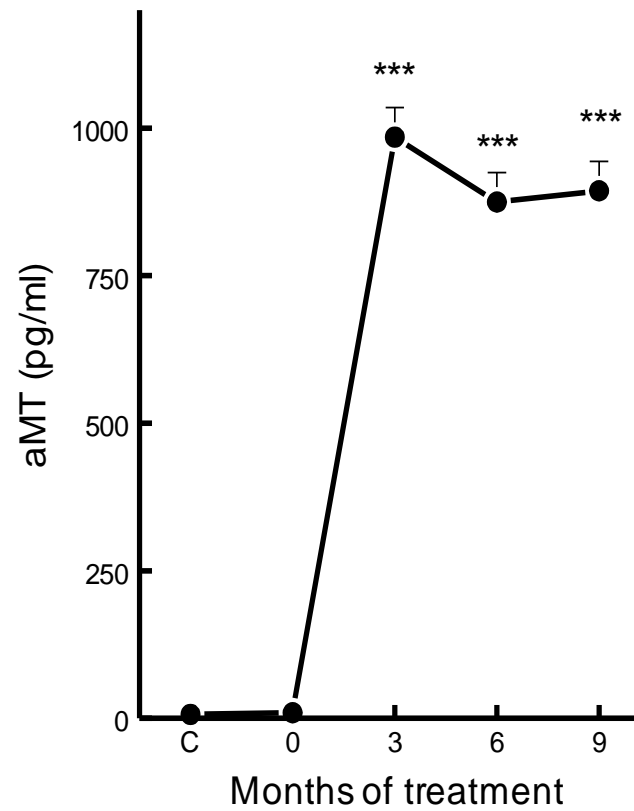
Biochemical Parameter	Control (n = 10)	Treatment			
		Time 0 (n = 10)	3 months (n = 9)	6 months (n = 7)	9 months (n = 5)
CK	38-190	7301 ± 2219	4912 ± 2007	6149 ± 1827	3969 ± 1617
AST	10-38	143 ± 37	110 ± 32	131 ± 26	103 ± 34
ALT	5-41	200 ± 52	170 ± 55	192 ± 39	159 ± 50
γGT	5-40	16 ± 2	15 ± 2	21 ± 2	17 ± 3
LDH	240-480	1362 ± 313	7881 ± 313	1119 ± 263	982 ± 325
Aldolase	3-12	27 ± 7	33 ± 13	27 ± 6	24 ± 8
Myoglobin	28-72	654 ± 127	450 ± 98	505 ± 96	----

Data are expressed as the means ± SEM. Enzyme activities are expressed in U/L, and myoglobin concentration in ng/L. CK, creatin kinase; AST, aspartate aminotransferase; ALT, alanine aminotransferase; γGT, gamma glutamyl transpeptidase; LDH, lactate dehydrogenase.



<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN





<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

CONCLUSION



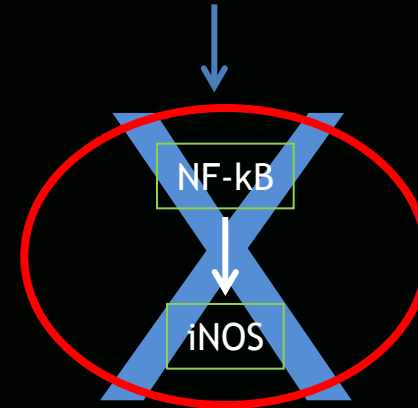
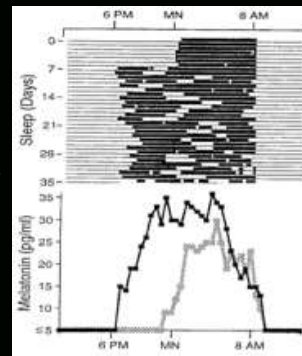
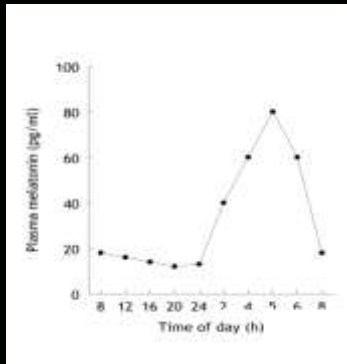
<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

MELATONIN

SYNCHRONIZES
THE
CIRCADIAN RHYTHMS

ANTIOXIDANTIVE AND
ANTIINFLAMMATORY
ACTIVITIES



DMD IMPROVEMENT



<http://melatonin.ugr.es>
dacuna@ugr.es

INSTITUTE OF BIOTECHNOLOGY
BIOMEDICAL RESEARCH CENTER
HEALTH SCIENCE TECHNOLOGY PARK
UNIVERSITY OF GRANADA, SPAIN

