dance that are associated with failure of electron transport function) have been demonstrated in the neurons of aged individuals and, in greater numbers, in Parkinson's Disease (PD) individuals.

Neuronal loss in the Locus Ceruleus, the major site of norepinephrine synthesis in the CNS, may exceed 50% in early AD and PD, leading some to speculate that a deficiency in this neurotransmitter system, possibly in combination with declines in other neurotransmitter systems, is a significant component of AD pathology. We have examined the neurons of the Locus Ceruleus in aged AD and non-AD subjects for evidence of electron transport chain failure and mitochondrial DNA deletion-mutations. We have found evidence for both, in a pattern similar to that observed in the substantia nigra in aged and PD individuals and in excess of that found in neurons from CNS regions not specialized for neurotransmitter synthesis.

We suggest that redox-based consequences of specific aspects of neurotransmitter synthesis and metabolism may put monoaminergic neurotransmitter neurons at particular risk for mitochondrial genetic damage, functional decline and eventual death during the course of a lifetime. The resulting relentless decline in multiple neurotransmitter systems may play a central role in fundamental and/or associated deficits in AD, PD and other primary neurodegenerative diseases.

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141 Mitochondrial transcription factor a mitochondrial DNA repair

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Mitochondrial transcription factor A (TFAM, previously known as mtTFA) is an essential component of mitochondrial nucleoids. TFAM's role as a key regulator of mitochondrial DNA transcription and replication is now established, however, it is unclear whether this protein is involved in mitochondrial DNA repair. The main purpose of this study was to characterize the role of TFAM in mitochondrial base excision repair (BER), the only complete biochemical pathway for oxidative mtDNA damage repair characterized so far in mitochondria. Survival assays after oxidative (H_20_2, Menadione) or alkylation (MMS) stress reveal that depletion of TFAM results in cells that are more resistant to all of these stressors. Recombinant human TFAM was produced in a bacterial system and purified. Binding studies showed that the presence of 8-oxoguanine, one of the most common oxidative damage increases TFAM binding to DNA significantly, while the presence of an abasic site, a uracil, or a gap in the sequence did not interfere with the binding observed. Activities of the different enzymes involved in the process of base excision repair were determined and showed that TFAM inhibited the incision activity of 8-oxoguanine DNA glycosylase, uracil-DNA glycosylase (UDG), apurinic endonuclease 1 (APE1), as well as the incorporation activity of DNA polymerase gamma in vitro. Currently, we are evaluating whether TFAM and mitochondrial BER proteins interact. All together, the results suggest that TFAM is likely to play a role in regulating the base excision repair process in mitochondria.

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142 Unraveling the potential role of human Suv3 in genome maintenance

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The *Saccharomyces cerevisiae* Suv3 helicase together with the nuclease Dss1 forms the degradosome complex, which plays a pivotal role in mitochondrial RNA metabolism, and consequently in mitochondrial homeostasis. Although evidence suggests that human Suv3 is a similar central player in orchestrating the post-transcriptional fate of mitochondrial RNA, its role might go beyond this functional niche. We have addressed the possible role of hSuv3 in genome maintenance by searching for interacting partners. By performing co-immunoprecipitations, we report an interaction in human cells between hSuv3 and the proteins Replication Protein A and Flap Endonuclease 1. Additionally, low amounts of RPA stimulate the helicase activity of hSuv3 while it is unaffected by a physical interaction with Fen1. On the other hand, the flap endonuclease activity of Fen1 is increased in the presence of hSuv3. In line with the recent discovery of Fen1 in the mitochondria, further studies are aimed at investigating potential implications of hSuv3 in mitochondrial BER. Current studies are aimed at the analysis of hSuv3's subcellular localization after DNA damage and its possible co-localization with RPA and Fen1 by indirect immunoflourescence. These results support extending the focus of the functional roles of hSuv3 to get a better understanding of the position of this essential protein in cell survival.

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143 Development of a cell based screening assay for drugs that alter mitochondrial function

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We describe a medium-to-high throughput screening method to search for drugs which enhance mitochondrial function in the context of the living cell. The method utilizes the Becton–Dickinson Oxygen Biosensor System, which employs fluorescence to measure cellular oxygen consumption. The assay works in several cell types, including neural, muscle, lymphoblast, osteosarcoma, HEK, and adipocyte, and detects deficits of function associated with mitochondrial mutations, mitochondrial disease and mitochondrial inhibitors. Edge effects were noted in the 96-well format, however baseline variability testing produced z' scores ranging from 0.39 to 0.76 in K562 cells using non-edge wells. Proof-of-principle was established for testing mitochondrial enhancers in the context of a partial rotenone block. Proof-of-principle was also demonstrated for the PGC1-alpha inducer AICAR in C2C12 muscle cells, and quantitative PCR demonstrated in mitochondrial to nuclear copy number ratio and in PGC1-alpha transcript. AICAR-treated cells had increased cellular ATP content. Overall, the methodology could be applied to screen available compound libraries for enhancers of mitochondrial function in the context of mitochondrial disease.

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144 Aberrant amino acid signalling and metabolism in the Deletor mouse

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We performed metabolomic and lipodomic profiling in the Deletor mouse, a disease model for the late-onset mitochondrial dis-