

Supplementary Material

The dual-site agonist for human M2 muscarinic receptors Iper-8-naphtalimide induces mitochondrial dysfunction in *Saccharomyces cerevisiae*.

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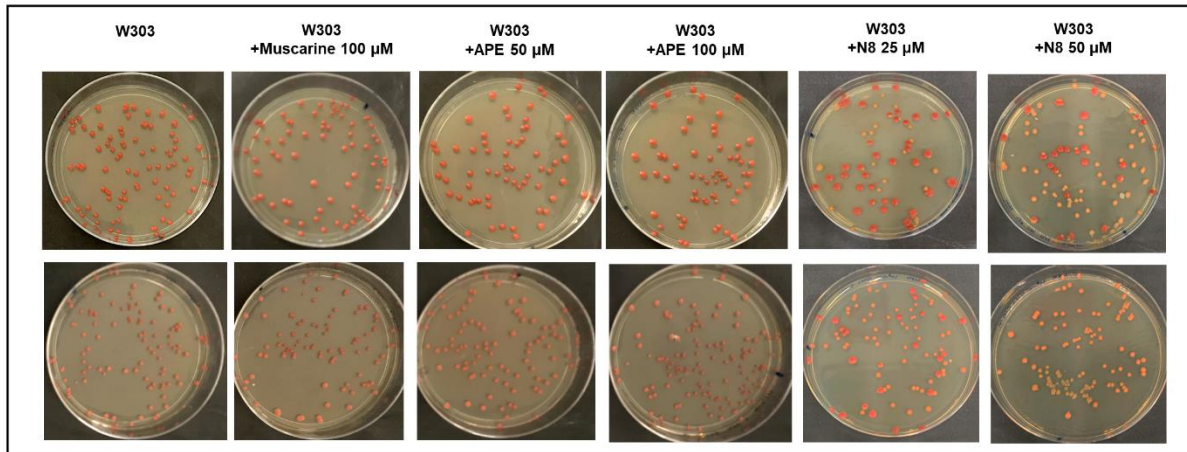


FIGURE S1: The muscarinic agonist N-8-Iper (N8) induces an high rate of petites production in yeast. Pictures show the growth of *S. cerevisiae* treated with muscarinic agonists in complete medium (YPD). Pictures were taken after 24 hours (upper pictures) and 48 hours (lower pictures) of growth. The agonist N-8-Iper respiration impairment is time and concentration dependent. Muscarine, the control, and APE have no effect on yeast cells mitochondrial function. The colours of the colonies facilitates the screening of colonies with a defect in respiration: the W303 wild type yeast colonies are red because the *ade2*- mutation results in the accumulation of the substrate of the Ade2, an enzyme of the adenine biosynthetic pathway. The formation of this red intermediate is produced into the mitochondria. If the mitochondria are not functional, the intermediate is not produced and the colonies result in white/brown colonies [31, 32].

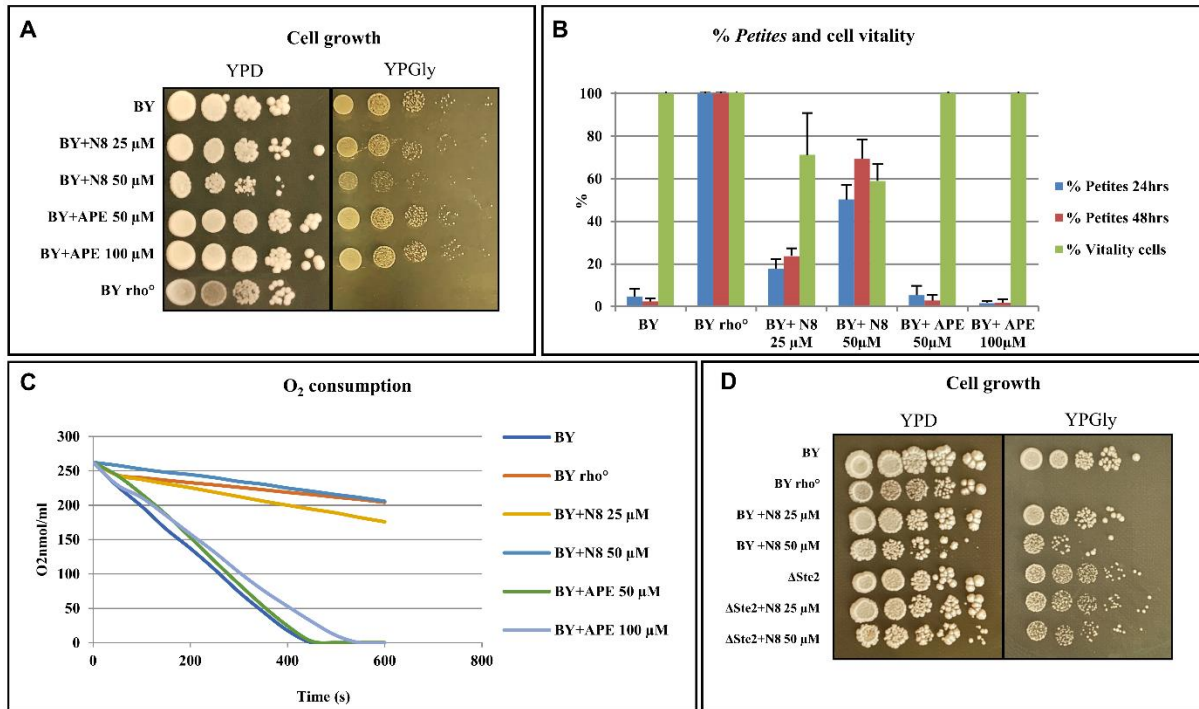


FIGURE S2: The muscarinic agonist N-8-Iper (N8) induces a mitochondrial defect in the *S. cerevisiae* BY4741 wild type strain. (A) Growth of BY4741 treated with muscarinic agonists in complete medium (YPD) and in a respiratory carbon source (YPGly), pictures taken after 24 hours of growth N-8-Iper impairs the mitochondrial function while APE has no effect. (B) Percentage of petites production and vitality of wild type BY4741, BY4741 rho^o grown in YPD medium and wild type BY4741 grown in YPD medium with agonists N-8-Iper (25 and 50 μ M) and APE (50 and 100 μ M). (C) The respiration rate of BY4741 yeast cells treated with N-8-Iper is impaired compared to the wild type strain, APE has no effect on the respiration rate. The oxygen consumption rate of BY4741, BY4741 rho^o and BY4741 cells treated with agonists is expressed in O₂nmol/ml. (D) Growth of BY4741 and Δ Ste2 treated with muscarinic agonists in complete medium (YPD) and in a respiratory carbon source (YPGly), pictures taken after 24 hours of growth N-8-Iper impairs the mitochondrial function.

REFERENCES

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