

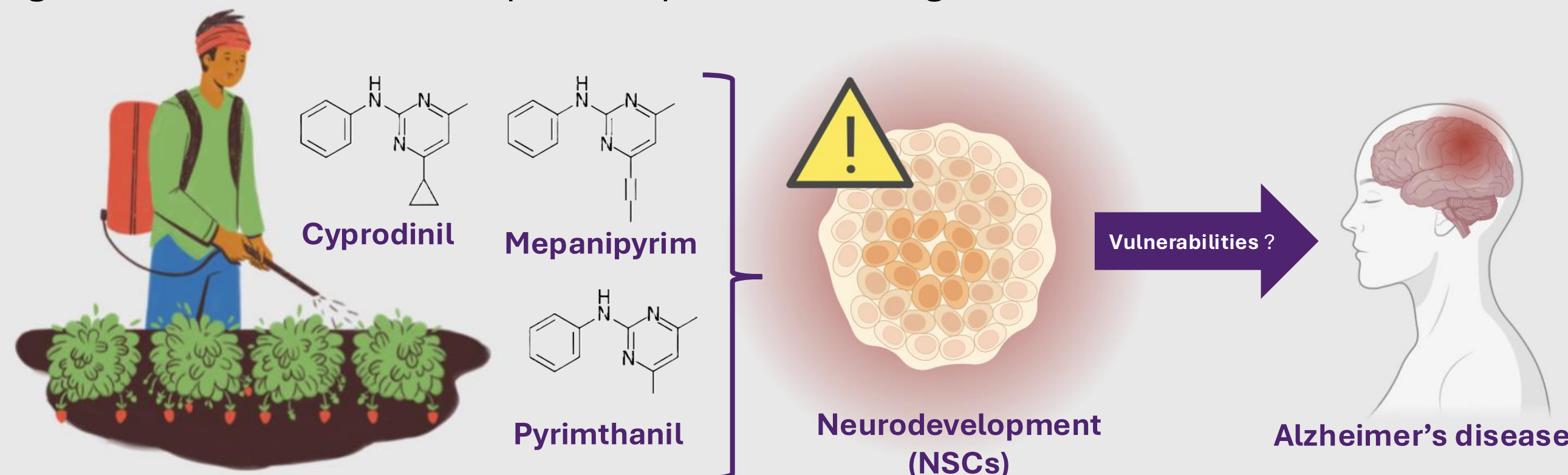
Neurodevelopmental Exposure To Fungicides : Potential Implications For Alzheimer's Disease Physiopathology

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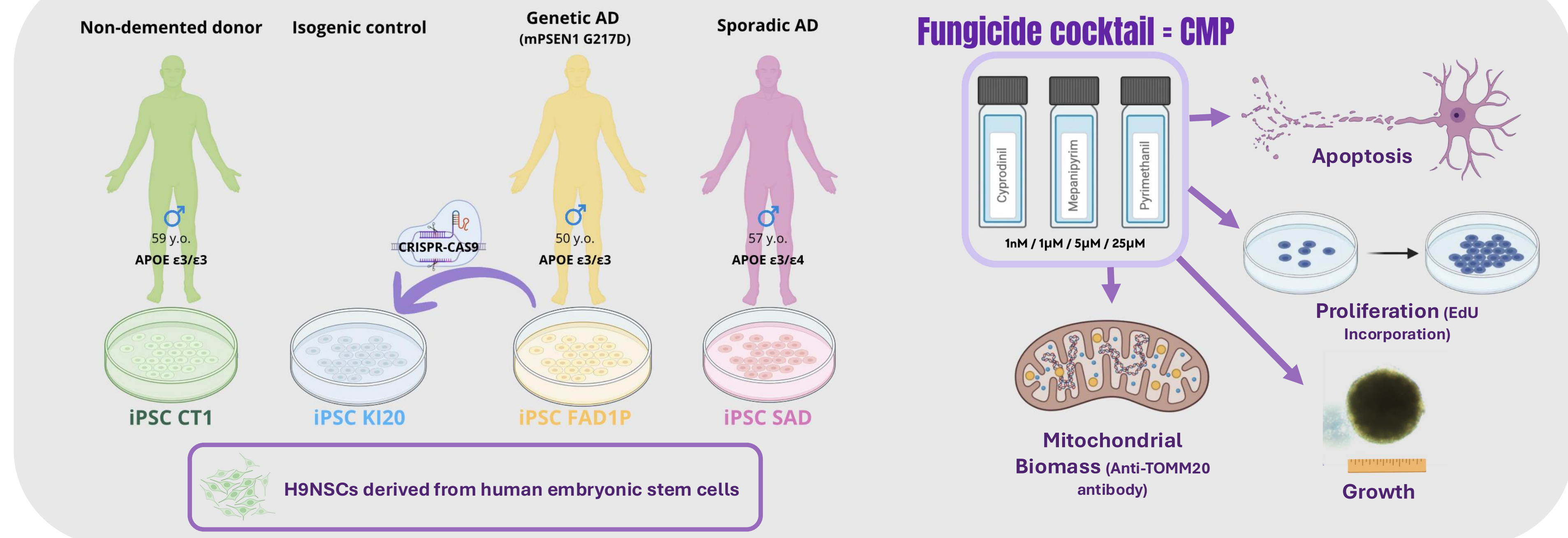
Exposure to anilinoypyrimidine fungicide cocktail disrupts key neural stem cells properties in human cellular models, including mitochondrial biomass, proliferation, apoptosis, and growth. These findings support the hypothesis that early-life pesticide exposure may alter neurodevelopment and increase vulnerability to neurodegenerative diseases such as Alzheimer's disease later in life.

INTRODUCTION

Neurodevelopment is a highly coordinated process involving neural stem cells (NSCs) proliferation, differentiation, migration and maturation. Increasing evidence suggests that environmental exposures occurring during **critical developmental windows** may induce **long-lasting cellular alterations** that could increase susceptibility to diseases later in life. We investigated the consequences of a fungicide exposure on organoid growth, cellular organization and neurodevelopmental processes using H9-NSCs and iPSCs.

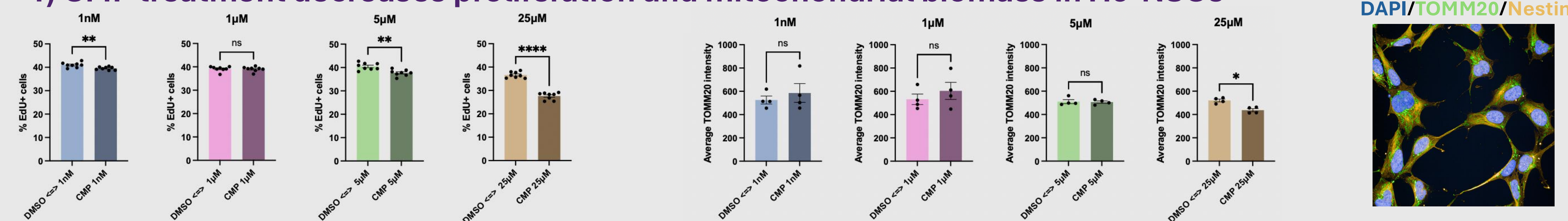


METHODS



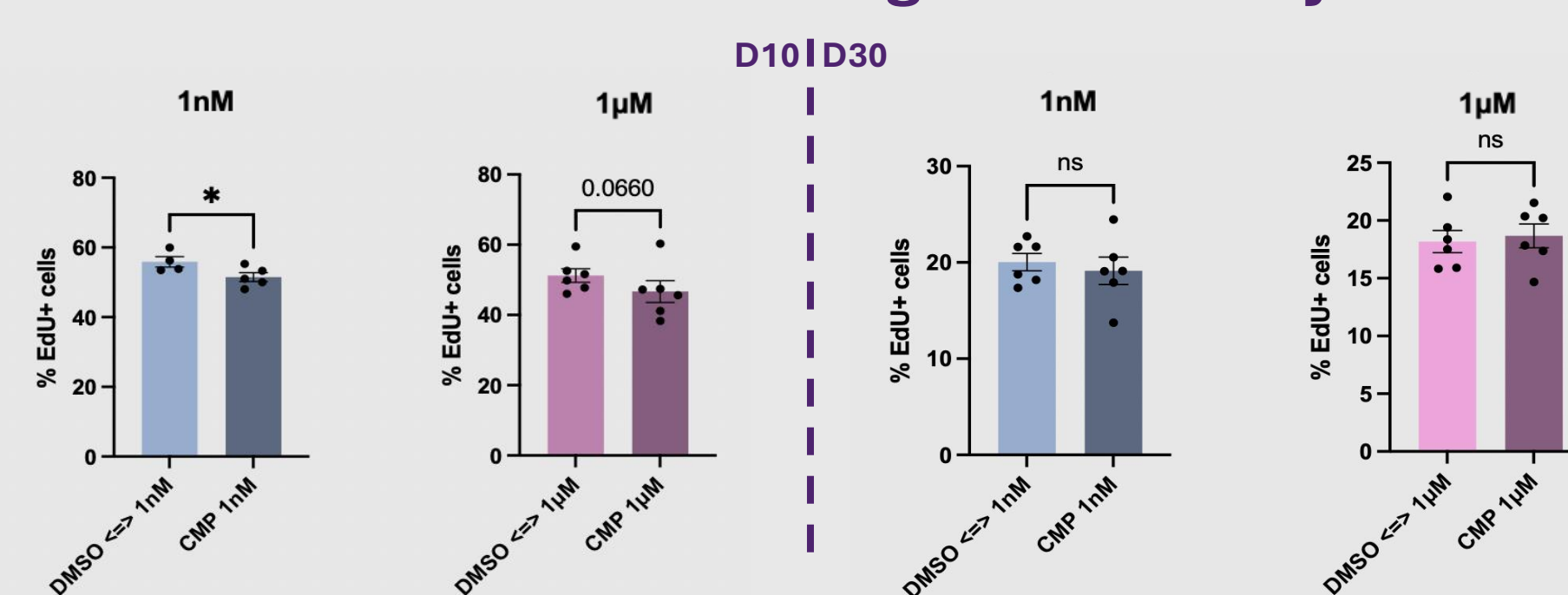
RESULTS

1) CMP treatment decreases proliferation and mitochondrial biomass in H9-NSCs



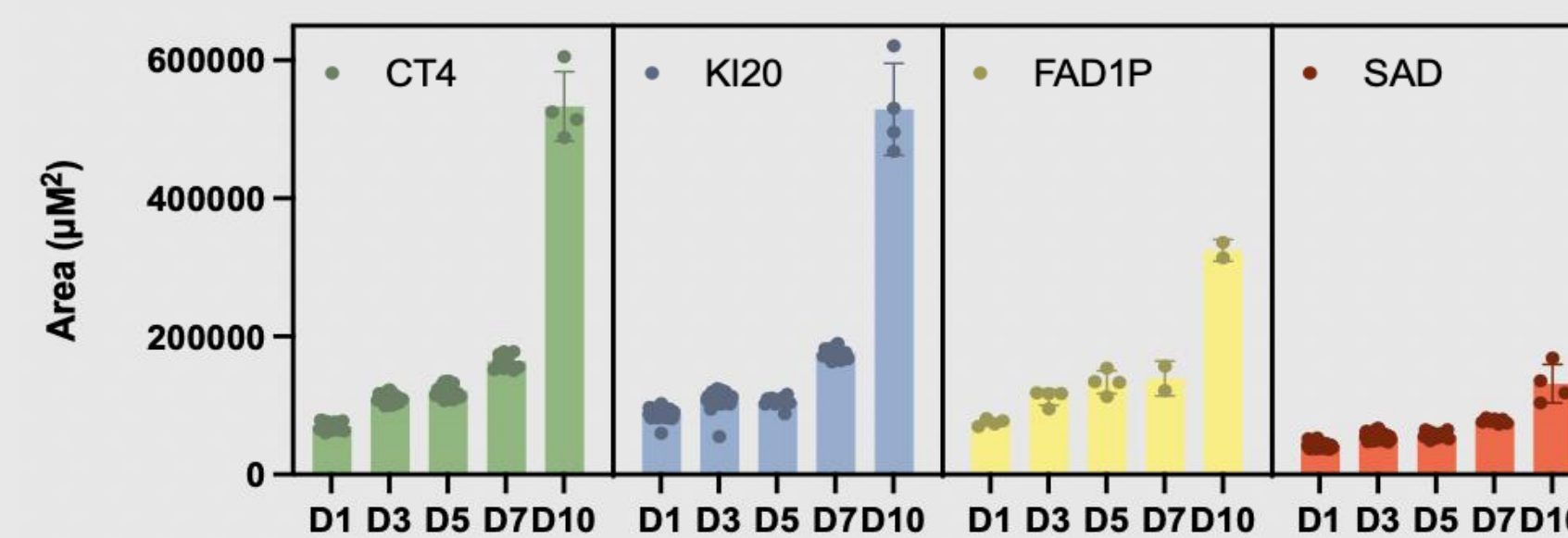
Fungicide cocktail exposure decreases proliferation and mitochondrial biomass in H9-NSCs : Cell proliferation was assessed by Edu incorporation following exposure to the CMP cocktail (1 nM, 1 μM, 5 μM or 25 μM) during 48h. Mitochondrial biomass was evaluated by quantifying TOMM20 fluorescence intensity in Nestin-positive H9-NSCs. Representative immunofluorescence staining (DAPI/TOMM20/Nestin) is shown. Results are presented as mean ± SEM. Statistical significance was determined using Mann-Whitney test. ns: not significant; *p<0.05; **p<0.01; ***p<0.001; ****p<0.0001.

2) CMP treatment decreases cell proliferation in non-demented cerebral organoids at day 10



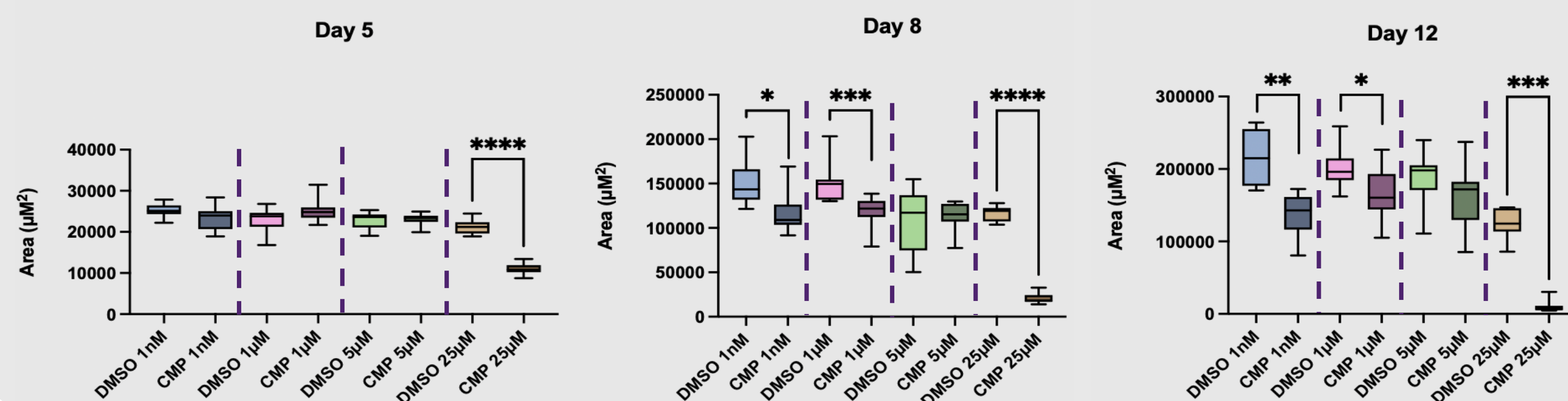
Fungicide cocktail exposure during neural induction reduces cell proliferation in non-demented organoids : Cell proliferation was assessed by Edu incorporation in non-demented (CT1) organoids exposed to the CMP cocktail for 30 days and cell proliferation was assessed at day 10 (D10) and day 30 (D30). Results are presented as mean ± SEM. Statistical significance was determined using Mann-Whitney test. ns: not significant; *p<0.05; **p<0.01; ***p<0.001; ****p<0.0001.

3) Individual and cellular properties influence Alzheimer's disease cerebral organoids' growth



Intrinsic cellular properties influence the growth of Alzheimer's disease organoids. The projected surface area of untreated organoids derived from CT4, KI20, FAD1P and SAD cell lines was measured throughout neural induction (days 1, 3, 5, 7 and 10). Results are presented as mean ± SEM, illustrating differences in growth kinetics between cell lines independent of CMP exposure.

4) CMP treatment alters the growth of non-demented cerebral organoids

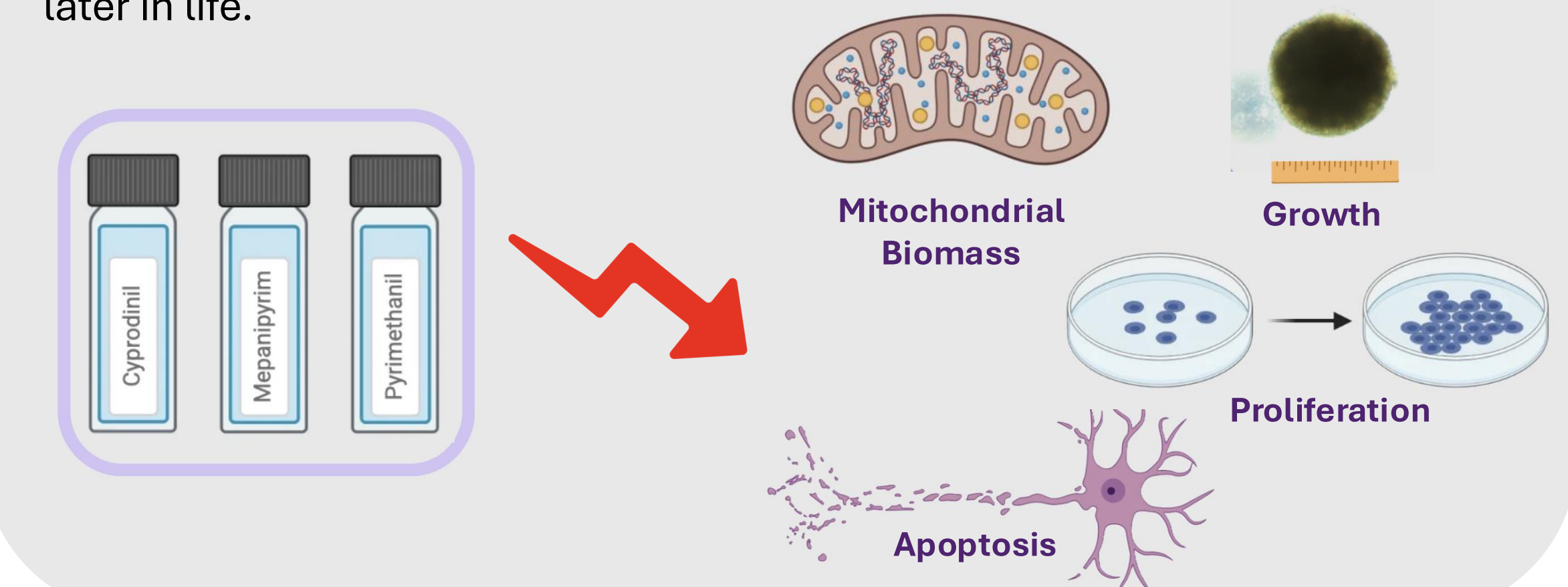


Fungicide cocktail exposure during neural induction impairs organoid growth : Organoid size was assessed by measuring the projected surface area of CMP cocktail-treated organoids compared with DMSO-treated organoids at day 5, day 8 and day 12 (6 ≤ n ≤ 12, Mann-Whitney).

Results are shown as box and whiskers plots (min to max). Asterisks : significant difference with DMSO group (t-test or Mann-Whitney); one symbol: p<0.05, two symbols: p<0.01; three symbols: p<0.001, four symbols: p<0.0001.

CONCLUSIONS

These findings support the hypothesis that **environmental pollutants** and particularly **anilinoypyrimidine fungicides**, can induce early cellular dysfunctions during brain development, potentially creating **vulnerabilities** that contribute to neurodegenerative diseases, such as Alzheimer's disease, later in life.



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