GASTROINTESTINAL DYSFUNCTION

Gastrointestinal (GI) dysfunction is a common issue in Parkinson’s disease (PD). Symptoms including dysphagia, bloating, and constipation, with constipation often reported decades before motor symptoms appear. Pathologically, alpha-synuclein (α-syn) accumulation has been reported in the neurons of the GI system, as well as morphology- and activity-based changes of the enteric nervous system (ENS), microbial dysbiosis, and increased gut permeability. Below you will find a summary of models that display GI dysfunction. Please note, this list is not comprehensive.

LEE G2-3 Prp-A53T SNCA MOUSE

- **Description:** This line displays overexpression of human A53T α-syn in brain and peripheral tissues. Motor deficits appear around 8-10 months of age and culminate in paralysis, but this is due to motor neuron loss rather than loss of dopamine neurons in the substantia nigra. When motor symptoms appear, robust astrogliosis and α-syn pathology are observed in the midbrain, brainstem, and spinal cord. Constipation and α-syn pathology in the colon are present at 3 months of age, prior to motor dysfunction and brain pathology. Interestingly, this line has been used in α-syn RT-QuIC experiments whereby colon tissues from 3 month old mice were able to demonstrate seeding activity prior to brain samples (brain seeding activity only at 12 months when motor symptoms were present).

- **Recommended Use:** Pathology in this model is driven by high levels of α-syn overexpression. The model is useful for studying synuclein pathology in the gut and resulting GI dysfunction that precedes synuclein pathology in the brain. As this model does not display nigrostriatal degeneration, it should not be used to evaluate that system.

- **Helpful Resources:**
  - Summary of Line Phenotypes – JPDN Model Summary
  - Commercial Availability – This model is available at JAX (JAX #006823).

NUSSBAUM PAC(SNCA A53T)+/+; Snca−/− MOUSE

- **Description:** This line displays overexpression of human A53T α-syn in brain and peripheral tissues without endogenous mouse α-syn present. Motor deficits are present beginning at 6 months. Synuclein pathology is not observed in this line and the nigrostriatal system remains intact with no nigral dopamine neuron loss or decrease in striatal dopamine levels. Decreased colonic motility is observed as early as 3 months of age with evidence of α-syn aggregation in ENS neurons.

- **Recommended Use:** Pathology in this model is driven by high levels of α-syn overexpression. The model is useful for studying synuclein pathology in the gut and resulting GI dysfunction, but should not be used in evaluating a link to brain pathology given the absence of nigrostriatal degeneration and lack of synuclein pathology in the nigra.

- **Helpful Resources:**
  - Summary of Line Phenotypes – JPDN Model Summary
  - Commercial Availability – This model is available at JAX (JAX #017099).

MASLIAH THY1 ASYN “LINE 61” MOUSE

- **Description:** This transgenic mouse model overexpresses human wildtype α-syn in the brain and peripheral tissues under the Thy1 promoter. The model displays robust α-syn pathology, primarily in the cortex and limbic system. Loss of dopaminergic terminals in the striatum occurs in this model but not until late timepoints. Inflammation, mitochondrial dysfunction, and motor phenotypes occur early. The line does not display loss of neurons in the substantia nigra. Reduced fecal output in response to novelty is observed at 3 months and impaired colonic motility is observed at 11-12 months of age. Synuclein is observed in the myenteric plexis around 8-10 months of age.
**MPTP MODEL**

- **Description:** MPTP is a toxin administered to mice that acts as a mitochondrial complex I inhibitor with high affinity for the dopamine transporter. There are different dosing paradigms for MPTP that result in different pathology:
  - The acute dosing paradigm (4 administrations in 24 hours) produces rapid loss of nigral dopamine neurons and decreased dopaminergic terminals in the striatum. Robust motor deficits occur but recover at longer post-injection intervals. αSyn pathology is not observed. GI dysfunction includes loss of dopamine neurons in the ENS and changes in gut microbiota. A transient increase in colonic motility 2-3 days after injection is observed, with constipation also reported at 7+ days post-injection (although not observed by all).
  - Sub-acute (1 administration daily for 5-7 days) and chronic (1 administration daily for 14-28 days) dosing paradigms result in delayed nigrostriatal degeneration with accompanying αSyn pathology and neuroinflammation. These paradigms do not result in robust motor deficits, as some report phenotypes while others do not. GI dysfunction includes changes in gut microbiota, increased gut permeability, and increased gut inflammation. αSyn pathology in the gut has been reported by many. Constipation and decreased dopamine in the ENS are not consistently reported.

- **Recommended Use:** The ability to administer MPTP through peripheral injection is a benefit of this model as it avoids the need for stereotaxic surgery. The robust CNS pathology and accompanying GI pathology are benefits but the continuation of conflicting reports on behavioral readouts of GI dysfunction remain a challenge. Note that mice and primates (including humans), but not rats, efficiently convert MPTP to its active metabolite, MPP+. Thus, rats are not compatible with this model and great care is imperative for the safe handling and disposal of MPTP.

- **Helpful Resources:**
  - CRO Recommendations for the MPTP Model - Atuka, Charles River Labs, Psychogenics

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**ROTENONE MODEL**

- **Description:** Rotenone is a toxic pesticide that inhibits mitochondrial complex 1 to produce degeneration when administered to mice and rats. Phenotypes vary based on route of administration and dose, but chronic, systemic administration results in loss of dopaminergic neurons in the substantia nigra, αSyn pathology, inflammation, and motor dysfunction. Regarding GI dysfunction, an early report using gastric administration of rotenone in mice claimed progressive inflammation and αSyn pathology beginning in the ENS and continuing to the brainstem and substantia nigra, leading to nigral dopaminergic neuron loss and motor phenotypes. Unfortunately, these phenotypes were not replicated in later studies. Subcutaneous/intraperitoneal administration to rats and mice appear to result in more robust gastroparesis, microbial dysbiosis, myenteric neuron loss, and gut αSyn pathology.

- **Recommended Use:** The nigrostriatal degeneration with αSyn pathology make this an attractive model. However, it should be noted that there is substantial variability in this model between different administration paradigms and even within a paradigm. Given the known toxicity of rotenone, great caution should be taken when handling this pesticide. For studies of GI dysfunction, systemic injection seems to be more reproducible than oral administration.

- **Helpful Resources:**
  - CRO Recommendations for the 6-OHDA Model - Atuka, Charles River Labs, Psychogenics
6-OHDA MODEL

- **Description:** 6-OHDA is a mitochondrial complex I and IV inhibitor that is administered through stereotaxic injection into the rat brain via the striatum, substantia nigra, or medial forebrain bundle (MFB). The route of administration affects pathology in this model – nigral or MFB injection will result in loss of nigral dopamine neurons followed by degeneration of striatal terminals within days, whereas striatal injection results in a progressive model that begins with degeneration of striatal dopamine terminals and results in loss of nigral dopamine neurons in a matter of weeks. Regardless of injected structure, robust unilateral motor deficits are present and neuroinflammation is observed. αSyn pathology, however, is not present in this model. Regarding GI dysfunction, unilateral injection of 6-OHDA to rat nigra results in increased colonic tyrosine hydroxylase and constipation. Bilateral injection of 6-OHDA to the rat nigra results in constipation, increased intestinal permeability, and gut inflammation.

- **Recommended Use:** 6-OHDA is a potent inhibitor of mitochondrial respiration complexes I and IV. The robust degeneration and motor deficits make this an attractive model. The robust CNS pathology and accompanying constipation phenotypes are attractive, but the exact mechanism of the GI dysfunction remains undetermined.

- **Helpful Resources:**
  - CRO Recommendations for the 6-OHDA Model - Atuka, Charles River Labs, Psychogenics

MITOPARK MOUSE MODEL

- **Description:** The MitoPark model involves homozygous knockout of Tfam specifically in midbrain dopaminergic neurons, resulting in impairments in mtDNA maintenance and the mitochondrial respiratory chain. This model displays progressive loss of dopaminergic neurons in the nigra starting at 3 months, corresponding to a progressive loss of striatal dopamine neuron terminals. Motor deficits appear at 3 months of age and continue throughout the lifespan. Synuclein inclusions are observed in dopamine neurons beginning at 1.5 months. Regarding GI dysfunction, this model displays early GI motility deficits that appear prior to motor symptoms. Additional phenotypes include constipation, gut inflammation, loss of dopamine neurons in the gut, and microbial dysbiosis.

- **Recommended Use:** This model is attractive in that GI dysfunction precedes motor impairment, much like the PD condition. However, it is important to note that the model does not necessarily reflect the pathogenic mechanisms of PD and may fail to recapitulate changes in other pathways that are important in human disease.

- **Helpful Resources:**
  - Commercial Availability – This model is not commercially available.

GASTRIC ASYN PFF MODEL

- **Description:** This model uses injection of recombinant aSyn aggregates that are 50nm or smaller (known as preformed fibrils or PFFs) into the mouse or rat gastric wall. Injection of these PFFs results in templating of the endogenous synuclein to induce pathological modifications. aSyn inclusions generally appear in myenteric ganglia and dorsal motor nucleus of the vagus. While some have reported additional pathology in ascending CNS regions, these readouts have been mired by conflicting reports. Constipation and inflammation have been reported.

- **Recommended Use:** This model is often chosen by groups seeking to study the possible gut-to-brain spread of aSyn. However, given the many conflicting reports on pathology, one should review literature as a whole and perform pilot studies before embarking on a large study using this model.

- **Helpful Resources:**
  - Commercial aSyn PFF sources – MJFF aSyn PFFs (sold as monomer) or StressMarq aSyn PFFs.
  - CRO Recommendations – Atuka, Psychogenics