NINDS PD2014: Final Research Recommendations

CLINICAL RESEARCH RECOMMENDATIONS (10)

***Recommendation 1:* Define the features and natural history of prodromal PD including progression, events that underlie phenoconversion to clinically manifest PD, and biomarkers or other determinants of prodromal subtypes with the goal of providing sufficient rationale to initiate proof-of-concept prevention trials that initially target high-risk populations.**

Need: PD has a preclinical phase, and by the time neurologic symptoms emerge, pathologic changes in the brain are widespread. Earlier detection and intervention are needed in order to optimally intervene in the PD process. The nature and duration of prodromal PD is largely unknown, but its characterization is critical to understanding the natural history of PD and its variable clinical phenotypes. Further, understanding the phase of PD before neurologic symptoms emerge will provide the foundation for PD-modifying and prevention clinical trials. Identifying at-risk cohorts who volunteer to contribute to a repository of biological and clinical data will be essential to establishing appropriate screening paradigms and early treatment strategies.

Approaches:

- Develop efficient screening paradigms to identify high-risk populations, *e.g.*, individuals with inherited PD or recognized prodromal symptoms such as REM Sleep Behavior Disorder, hyposmia, or novel measures of increased risk, before significant neurodegeneration has occurred.
- Conduct studies in individuals with prodromal markers to determine the risk for progression of these markers and also the risk of transition to clinically manifest PD. These studies also would lead to consensus around a research definition of phenoconversion to clinically manifest PD.
- Initiate biomarker studies including genetics, cerebrospinal fluid (CSF), imaging, proteomics, and metabolomics to correlate with PD risk.
- Enable the start of PD prevention trials. Such trials would integrate biomarkers to generate robust measures of PD pathophysiologic processes, target engagement, and downstream biological effects. They would also generate a database of publicly available results and tissue repositories, including autopsy material that can be used to accelerate therapeutic development and understanding of PD.

***Recommendation 2:* Develop effective treatments and companion biomarkers for dopa-resistant features of PD. These features include both motor symptoms, particularly gait and balance problems such as freezing of gait, and non-motor symptoms, especially cognitive impairment, psychosis, and dysautonomia.**

Need: Non-motor symptoms and levodopa-resistant motor features collectively constitute a major area of therapeutic need in PD. Both non-motor symptoms and levodopa-resistant features, particularly postural instability, falls, and freezing of gait, are associated with significant morbidity and mortality, particularly in advanced PD. Non-motor symptoms of PD cover a broad clinical spectrum including cognitive impairment, psychosis, and autonomic failure and thus contribute to the clinical heterogeneity of the disease. The

underlying pathophysiologic mechanisms and progression of both non-motor symptoms and levodopa-resistant motor symptoms are driven by non-dopaminergic mechanisms and are poorly understood.

In addition, there is currently no consensus on the range of non-motor symptoms that should be systematically documented and the instruments that should be used. In striking contrast to the prevalence and clinical impact of non-motor symptoms, very few randomized clinical trials have been conducted to specifically target non-motor symptoms. Identification of PD subgroups by risk for specific non-motor symptoms and development of novel interventions for treatment or prevention of non-motor symptoms would result in improved quality of life for a majority of patients with PD.

Similarly, substantial knowledge gaps remain in relation to levodopa-resistant motor symptoms, particularly gait and balance problems. Recent research has hypothesized that dysfunctional cholinergic neurons in the pedunculopontine nucleus may play a role, but attempts to modulate their activity through targeted DBS or drug therapies have been disappointing. There is an urgent need to better understand the underlying mechanisms and clinic-pathologic correlations for these symptoms.

Approaches:

- Develop biomarkers, *e.g*., neurophysiological, imaging, and tissue, associated with specific non-motor symptoms or dopa-resistant disorders of gait and balance to identify patient subgroups with different clinical trajectories and to assess response to treatment.
- Use patient- and caregiver-derived information to prioritize investigation of non-motor symptoms and systematically use validated instruments to determine the most sensitive and specific outcome measures for each non-motor symptom.
- Define dysfunctional motor patterns in patients with gait and balance problems using body fixed sensors and other novel computational technology.
- Initiate clinical trials for non-motor symptoms using existing and newly developed symptomatic therapies that address key symptoms, such as dementia and psychosis, which impact patient function and the burden on caregivers.
- Identify novel targets for DBS and pharmacological therapies and study the role of non-pharmacologic approaches to treat gait and balance problems in advanced PD.

<u>Recommendation 3:</u> Characterize the long-term progression of PD and understand the mechanisms that underlie its heterogeneity in clinical presentation and rates of progression. Factors related to disease heterogeneity may include clusters of clinical features as well as biological factors such as genotype and biomarkers.

Need: Despite the benefits of dopaminergic therapies, improvement of signs and symptoms is temporary in the setting of progressive disabilities such as cognitive impairment, psychosis, postural instability, failing speech/swallowing. A more detailed understanding of risk for more rapidly progressive disability would facilitate development of treatments intended to improve long-term outcomes for PD patients. Clinical features and biomarkers could be used to predict risk for faster progression and also to identify relatively homogeneous patient populations that may be more likely to respond to targeted therapies. In particular, there is a need for biomarkers of disease activity based on specific molecular targets, *e.g*., α-synuclein or amyloid-beta (Aβ), involved in PD pathogenesis both to predict progression and to measure response to treatment.

Approaches:

- Conduct studies in well characterized PD cohorts followed over long time horizons (at least 5-10 years or greater) to identify features and associated biologic or genetic markers of progressive disability and

risk for specific motor and non-motor outcomes. Long-term outcomes may be assessed efficiently using technologies such as electronic medical records or telemedicine.
- Provide an accessible resource that enables sharing of de-identified clinical data, biomarkers, and genotypes that will amplify the impact of the knowledge that emerges from these long-term studies.
- Use longitudinal cohort data to perform clinical trial simulations to help determine optimal trial designs for treatments intended to reduce long-term disability or prevent motor or non-motor outcomes.
- Validate biomarkers of disease activity and target engagement to predict progression of motor and non-motor features and measure response to treatment.

<u>Recommendation 4:</u> Develop biomarkers of target engagement and proximal pharmacodynamic effects for use in early-stage clinical trials.

Need: In addition to safety and pharmacokinetics, early stage trials seek to determine if the experimental agent has engaged the intended biological target and had the appropriate pharmacological effect. Some of these markers may also be useful to enrich the study cohort to ensure that those subjects express the biological target at sufficient levels, *e.g*., amyloid imaging. Such information is critical for determining dose and regimen and supporting longer-term studies to test clinical efficacy. In the absence of these markers, it is not possible to know if the biological hypothesis was tested. Examples of targets in PD in brain tissue, other tissue, or fluids include but are not limited to α-synuclein, GBA, LRRK2, and parkin. Targets such as Aβ and tau may overlap with other neurodegenerative diseases.

Approaches:

- Coordinate focused efforts to develop imaging, *e.g*., α-synuclein imaging agent, or other assays for a limited set of genetically defined targets that are likely to be key to disease modification. These are different from progression markers and need not have longitudinal follow-up.
- Employ a variety of different *in vitro* and *in vivo* approaches to identify relevant markers.
- Conduct focused efforts to study evolving biomarkers from AD and related neurodegenerative disorders (such as Aβ and tau imaging ligands) to determine their association with features of PD such as cognitive impairment.

<u>Recommendation 5:</u> Improve methods to assess long-term efficacy and potential for disease modification in clinical trials, including more efficient strategies for screening potential agent and trial design simulations to assess the performance for predicting long-term benefits.

Need: Since confirmatory trials of disease modification require long-term follow-up, efficient designs to screen potential agents are needed for PD, especially approaches that allow assessing more than one treatment or multiple dosages of the same treatment at the same time. Currently, when a treatment fails to show efficacy in a clinical trial, it is often not clear whether the failure was due solely to the treatment itself or also to some deficiency in the trial design or measurement of the disease implemented. As novel methods and approaches are developed, it will be important to examine the performance of the design in situations where the "truth" is known, *i.e*., an intervention is truly effective or not. Accomplishing this will require groups of clinicians and statisticians working collaboratively to develop and assess potential design strategies, independent of the implementation of any specific trial.

Approaches:

- Investigate novel design paradigms that efficiently and quickly identify long-term treatment benefits.
- Develop improved learning phase study designs to identify promising compounds with a high degree of sensitivity and specificity, thus addressing limitations of existing strategies such as futility trials.

- Conduct simulation studies led by multi-disciplinary teams of clinical trialists to identify optimal trial designs.

<u>Recommendation 6:</u> Determine factors that facilitate public health interventions, including risk factor reduction and health services interventions for populations and individuals.

Need: The number of PD cases in the US will continue to increase with population aging, causing a huge social and economic burden; preventing PD is critical. PD is a complex disorder, with genetic and environmental determinants, providing an opportunity for identification of at-risk persons and population-wide risk reduction. PD has a long preclinical phase, providing a window of opportunity for disease-modifying interventions. Knowledge gaps that must be addressed include understanding risk and preventative factors, characteristics of latent and prodromal states, and determinants of disease progression. This knowledge is needed to develop interventions to prevent onset, or slow or stop progression.

Approaches:

- Investigate PD "at risk" and control populations to identify risk and preventative factors.
- Characterize preclinical PD features and the natural history of prodromal disease progression.
- Determine an efficient population screening method to identify those people for whom preventive interventions are indicated.
- Develop methods for implementing preventive interventions and assessing their efficacy.

<u>Recommendation 7:</u> Use of innovative outcome measures to evaluate motor and non-motor features, including patient- and clinician-reported outcomes that leverage emerging information technology (IT) opportunities, enhance sensitivity and specificity of measurement, and facilitate long-term follow-up of well-characterized cohorts.

Need: Outcomes measurement is a cornerstone of clinical research. Limitations in outcomes measurement serve as a "common denominator" resulting in limitations across the breadth of clinical research. Our understanding of PD has expanded to include motor and non-motor manifestations with greater insight into disability associated with individual symptoms, such as falls, fatigue, freezing, and cognitive impairment. However, high-quality tools for assessment of these diverse symptoms have not been adequately investigated. Modern measurement principles provide new opportunities to improve the quality of applied outcome measures including improved sensitivity, specificity, and practicality with the use of emerging technologies.

Approaches:

- Perform comparative studies of the sensitivity and specificity of available instruments including NIH NeuroQoL, PROMIS, Toolbox and Common Data Elements, with a focus on the optimum application of patient- and clinician-reported outcomes and physical/cognitive performance measures.
- Investigate the magnitude of clinically important differences on a range of patient- and clinician-reported outcomes and physical/cognitive performance measures.
- Investigate the comparative feasibility and validity of diverse methods of data collection including computerized tablet, smartphone, remote monitoring, and computerized adaptive testing.
- Investigate the potential to use electronic medical records or telemedicine to capture long-term outcomes in clinical research cohorts.

<u>Recommendation 8:</u> Develop improved informatics capability to include investigation of how "big data" may contribute to a fuller understanding of PD, a central repository for PD trial data, a resource for trial design simulations.

Need: A more detailed understanding of the natural history of PD, both before and after diagnosis, and the characterization of PD subtypes would accelerate development of effective treatments. Making use of existing datasets is an efficient way to address these questions. Moreover, the design of clinical trials needs access to existing longitudinal data to model appropriately and simulate design operating characteristics. Although there are recommended common data elements and requirements for sharing data publically, the technical aspects of combining datasets across multiple sources require a considerable investment of time and expertise. PD trials and well-designed cohort studies provide the highest quality data and are a resource that needs to be preserved. Administrative data sources, such as electronic medical records, are a growing resource, but require planning to utilize.

Approaches:

- Maintain a central repository to standardize and uniformly archive existing and future trial data to maximize the usefulness of existing datasets.
- Encourage a central body to manage the standardization of datasets and provide a user-friendly end product. This resource would ensure that existing trial data are used appropriately to answer questions beyond the original intent for which they were collected. This central body would likely involve an ongoing team of data managers, programmers, and statisticians with the appropriate expertise in this area.
- Support the development of informatics to archive administrative data sources and explore ways to overcome barriers to use, such as access, ethical challenges, de-identification of data, and common data items.

<u>Recommendation 9:</u> Develop strategies to increase minority participation in research. These initiatives should include mechanisms to assess the effectiveness of these programs and could lead to the establishment of shared resources to facilitate minority recruitment in PD clinical trials.

Need: Members of ethnic and racial minorities have historically been underrepresented in PD research, and participation is substantially lower for PD than for other neurological disorders such as AD and stroke. To date, the specific barriers to minority research participation have not been successfully addressed. Greater minority participation would provide a basis to understand possible biological differences in the expression of PD and give confidence in the generalizability of research results to these populations.

Approaches:

- Undertake studies to address biological and clinical differences in the expression of PD in minority populations.
- Identify specific barriers to greater minority participation in PD research studies and develop actionable and measurable policy guidelines and recommendations for recruitment, enrollment, and retention of minorities in PD research.
- Undertake demonstration programs to test strategies that facilitate minority participation in PD research. These programs should include clearly defined plans to assess their effectiveness.
- Develop a best practices toolkit and other shared resources targeted toward community members, researchers, and government. These resources could include:
    - Rosters of local and regional research champions in minority populations
    - Training program for researchers on effective strategies to recruit, enroll, and retain minority participants
    - Access to infrastructure belonging to NINDS and other federal research programs that have successfully included minority populations
    - Initiatives to defray costs related to minority recruitment and retention

<u>Recommendation 10:</u> Identify risk factors and pathogenic mechanisms of motor fluctuations and dyskinesias to identify novel targets for prevention and symptomatic therapy.

Need: After chronic levodopa therapy in PD, patients develop a stereotyped pattern of motor dysfunction in which they cycle between an effectively medicated "on" state with good mobility, and an un-medicated, immobile state, in spite of frequent medication dosing and the use of levodopa extenders such as catechol-O-methyl transferase (COMT) inhibitors and slow release formulations. Excess involuntary movements, or dyskinesias often accompany the "on" state. Currently there are several symptomatic treatments for fluctuations and dyskinesias, including DBS and, potentially, continuous intestinal infusion of duodopa. The knowledge gaps in this area include the neural basis for fluctuations and dyskinesias, the neural basis for the efficacy of DBS, and the risk factors for development. A better understanding of these mechanisms will improve current symptomatic therapies and perhaps slow onset of fluctuations and dyskinesias in individuals identified as having highest risk.

Approaches:

- Develop novel neurophysiological and imaging tools to understand neural networks responsible for the development of motor fluctuations and dyskinesias.
- Identify genetic and other biomarkers for motor complication risk.
- Define novel targets for symptomatic therapies of motor complications.

TRANSLATIONAL RESEARCH RECOMMENDATIONS (10)

***Recommendation 1*: Develop patient stratification tools that define disease signatures of more homogeneous cohorts with emphasis on slow vs. fast progressing PD, prodromal PD, and non-motor symptoms.**

Need: PD is a clinical diagnosis but there exists a need to supplement this clinical classification in order to improve sensitivity and specificity. This is further confounded, as most patients do not have an identifiable cause that can be attributed to their diagnosis. If PD indeed has different causes, then therapeutic strategies are needed that enable the selection of patients who are most likely to benefit from the intended therapeutic. Current clinical trials are powered to detect effects assuming the majority of enrolled patients are responsive, so if only a subset would potentially benefit from the test agent, the clinical trial will fail even if the agent is effective in a subset of patients.

Approaches:

- Identify the most promising biomarkers for patient stratification.
- Develop sensitive imaging and biofluid assays for α-synuclein to assess LB accumulation and soluble species of α-synuclein.
- Develop novel genetic and genomic markers, markers of neuron health, and synaptic proteins
- Develop an understanding of the factors contributing to heterogeneity in the rate of progression of PD and the apparent response to certain interventions.
- Use currently available high-resolution metabolomics tools to analyze blood, urine, and CSF samples from extensively phenotyped PD patients to determine metabolic sub-classifications.
- Utilize genome-wide association study (GWAS) data, which have provided useful characterization of genetic risk factors for PD. Studies evaluating metabolic interactions with risk alleles are likely to provide new insight into disease mechanisms and potential for new therapeutic targets, especially for prevention or delay of progression.

***Recommendation 2*: Develop novel and specific PET imaging agents and assays to measure α-synuclein burden, validated in both animal models and human tissue.**

Need: Definitive non-invasive confirmation of pathologic α-synuclein is critical to support the accuracy of clinical diagnosis and can be used in combination with CSF measures of α-synuclein, to track disease progression and to monitor the effect of targeted therapeutics. Improvements or changes in α-synuclein levels may result not only from therapies targeted directly at α-synuclein but also from other successful therapies. Measurement of multiple α-synuclein species can be used as a pharmacodynamic marker.

Approaches:

- Develop selective and potent α-synuclein tracers analogous to the strategies used for Aβ and PHF-tau. Following structure-activity trends, maximizing α-synuclein selectivity and minimizing binding to other amyloid proteins can attain selectivity for α-synuclein over other amyloid proteins. α-synuclein, being a pathological hallmark of PD, much like Aβ and tau are pathological hallmarks of AD, may serve as a diagnostic target for PD diagnosis and for monitoring disease progression.

- Develop validated and standardized assay methodologies to measure soluble, oligomeric, and post-translational modified forms of α-synuclein in plasma and CSF.

<u>Recommendation 3</u>: Develop translational resources with greater power to predict efficacy and biomarker outcomes in clinical trials. These resources would include well-characterized, replication sets of iPS cell lines from sporadic, dominant, and recessive PD cases.

Need: To move the PD field forward we need a better understanding of the underlying basic mechanisms of disease, translation of these mechanisms into potential therapies, and cutting-edge clinical trials. Because of the poor track record of translating results from preclinical rodent toxin models to results in humans, the relevance of mechanisms discovered with these models to PD has been questioned and the investment by industry in developing therapies for PD is less than it might be. Thus, an important goal will be to create tools and models that can be used to study PD biology in a human context and in turn yield results that provide more accurate guidance for PD therapeutic development.

Approaches:

- Integrate iPS cell technology into all translational efforts for PD. To accomplish this goal, there should be well-characterized sets of non-integrating lines available from 10 sporadic cases and 10 cases from patients with each major PD gene mutation. Each mutant line should have a matching isogenic control corrected using gene targeting. iPS cell lines should be derived from patients with extensive clinical characterization and whose contributed biosamples have been used in "omic" studies. These lines should be available as frozen suspensions of neural progenitors specified to either a forebrain or hindbrain fate that can simply be thawed and plated by the PD researcher for use in their studies.
- Support predictive and interactive studies that inform relevant clinical PD phenotypes such as dopamine biology, pesticide sensitivity, etc. and that can provide readouts which can in turn be provided to clinical studies to help establish a relationship between cellular readouts and patient phenotypes.
- Encourage research supporting innovation that drives maturation and enhanced complexity of cell-based assay systems (e.g. 3-D systems, nigral/striatal co-cultures, transplantation of cells into immune deficient mice).
- Validate the utility of iPS cell models for providing information that predicts clinical findings. One approach would be to use systems biological and computational approaches to determine which iPS cell phenotypes, if any, correlate to clinical features of the patient from whom the cells came. If some sets of iPS cell phenotypes proved to predict clinical progression, for example, this type of analysis could be used in the future to stratify patients for a clinical trial. Similarly, efforts could be made in industry to retain tissue samples from patients enrolled in a clinical trial that could be used to make iPS cells selectively from responders and non-responders to determine whether the results for specific patients in the clinical trial could be predicted from the responses of their iPS cells.
- The likelihood of new mechanistic insights from toxin models of PD seems low, given the extent to which they have already been characterized and the limited power these models have had for predicting clinical trial results. Thus, the development of new animal models of PD should emphasize genetic causes or transmissibility mechanisms where the likelihood of novel mechanistic insights is higher.

<u>Recommendation 4</u>: Develop an integrated PD knowledge base that includes data from genetic, biomarker, clinical research, and clinical trials with informatics support for integration of existing databases for PD and other chronic neurodegenerative diseases.

Need: An enabling approach would be to develop PD signatures by collecting high-dimensional molecular data (*e.g.*, whole genome sequencing, transcriptomics, epigenomics, metabolomics, proteomics, iPS cell phenotypic data), and apply systems approaches to determine whether signatures of PD emerge that have important predictive value for clinical features, such as onset, progression, symptom profile and/or can identify new pathways and targets implicated in the pathophysiology of the disease. It is anticipated that disease signatures developed at different times would reveal whether the abnormalities found in patients cluster into a single group or multiple groups. Samples from clinical trials in PD should also be mined using the approaches described above to determine the extent to which responses among different PD patients are homogeneous or heterogeneous.

Approaches:

- Deploy systems approaches ("omics") on their own and in combination, especially using human information to develop an understanding of disease and effects of perturbations.
- Develop computational tools such as machine learning and other approaches to integrate disparate types of data into a global expression of disease signatures and to investigate the prognostic value of multidimensional data sets for disease progression (now) and response to therapy (longer-term).
- Support open access databases and shared resources

Recommendation 5: Establish consensus guidelines for preclinical therapeutic studies targeting α-synuclein to ensure appropriate use of existing models, improve replication of results across labs, and provide recommendations for future model development.

Need: Development of a standard set of models and procedures will facilitate direct comparison of various preclinical studies within labs and across different labs and treatment options. For example, in the case of the α-synuclein based models, there is a confusing array of possible models with very different outcome measures.

Approaches:

- Identify a few models that might be predictive for clinical trials and the appropriate outcome measures that should be used.
- If non-standard models are used, define appropriate outcome measures for targeting α-synuclein.
- Determine if α-synuclein causes common abnormalities in different models (transgenic, inoculation-spreading, virus-induced, and cell-based).
- Support standardized collection of pathologic tissue from animal models and people for unbiased pathway analysis.
- Organize an NINDS-sponsored workshop/panel to develop consensus guidelines for preclinical use of animal models for the assessment of *in vivo* assays. These guidelines should be reviewed regularly and modified.

Recommendation 6: Develop intermediate markers of drug efficacy in early PD translational studies to support more efficient proof-of-concept studies.

Need: Current clinical trials for disease modification in PD are expensive, require long periods of study and large numbers of patients, and future investment in such trials, and indeed in PD therapeutic development, is jeopardized due to the lack of previous success, and the finite resources within the overall funding system. Intermediate markers of drug efficacy could support shorter and more cost-effective proof-of-concept studies and ensure a continued investment in therapeutic development in PD.

Approaches:

- Investigate the use of neuroimaging biomarkers such as diffusion tensor imaging (DTI) or connectivity maps generated using MRI or blood-oxygen-level dependent (BOLD) MRI to assess their utility in shorter-term (*e.g.*, 6 months) trials.
- Study and validate neurophysiologic markers such as beta oscillations assessed by EEG/MEG in longitudinal trials to qualify their use in early trials.
- Explore studies that measure vesicular transport deficits in PD using CSF proteomics approaches in conjunction with labeling of the proteins via deuterated water ingestion. For therapies targeting α-synuclein, this functional end point might provide a measure of efficacy at a shorter time point.
- Support studies that will define the turnover rate of α-synuclein at different stages of PD. For instance, leucine labeling studies, as the ones conducted by at Washington University in St. Louis for Aβ, might provide a method to assess α-synuclein dynamics.

***Recommendation 7*: Define the required attributes of targets emerging from basic science efforts that justify advancement into translational studies in PD.**

Need: There are examples of failed high profile clinical studies of compounds in PD that have not been firmly grounded in a solid understanding of the mechanism of action of the compound tested as the intended therapeutic, or in some cases its molecular mechanism of action, and such compounds have advanced to clinical studies without any attempt at early translational studies such as target engagement or pharmacodynamics markers of drug action. There is a need to stop investment in such compounds in favor of those targets and compounds that are amenable to more rigorous translational approaches.

Approaches:

- Focus on targets that are firmly grounded in mechanistic studies and provide reproducible readouts in cell-based systems (*e.g.*, iPS-derived neurons or glia)
- Prioritize targets that can be detected and modulated *in vivo* with therapeutic approaches (*e.g*. enzyme activity in accessible peripheral compartments or CNS)
- Consider targets that are already "validated" in patients via genetic linkages between carriers of recessive genes and risk for common pathologies (*e.g*. *GBA* and PD)
- Insist on the development of target engagement biomarkers or companion diagnostics before a clinical trial is initiated to ensure that the results regarding the validity of the target are conclusive, whether the trial outcome is positive or negative.

***Recommendation 8*: Develop a thorough understanding of targets and pathways associated with pathogenesis and pathophysiologic mechanisms of PD with emphasis on those validated by human genetics and biology.**

Need: There is a need to agree on the most compelling and key pathways that are emerging in PD and have a strong foundation in human genetics or human biology, and to focus rigorous efforts to determine the most promising of these for future therapeutic development. At the same time there is a need to identify new promising targets that have not yet been identified.

Approaches:

- Confirm the validity of targets and pathways using human derived information such as genetics, tissue, and fluid "omics" approaches.
- Develop a well-curated biobank including brain tissue, CSF, blood, plasma, DNA, skin, *etc*.
- Determine whether there are convergent pathway(s) between dominant and recessive PD and whether this is contributing to sporadic PD.

- Deploy genetic screens in *Caenorhabditis elegans* or *Drosophila melanogaster* for enhancers and suppressors of genes already linked to PD to reveal new genetic steps in the known PD pathways and identify new drug targets. Complementary siRNA screens for genes in mammalian cells in defined PD pathways and cell based RNAi screens assessing wild type or mutant α-synuclein expression, aggregation, or autophagic engulfment might also be explored. Chemical genomic strategies stemming from cell based phenotypic drug screens could yield new drug targets following identification of the chemical targets.

<u>Recommendation 9</u>: Investigate the relationship between converging pathways in PD, for example α-synuclein misfolding and mitochondrial function.

Need: There is strong evidence for both α-synuclein misfolding and mitochondrial dysfunction in PD. One key question for translational drug development is whether these are unrelated causes of PD or if these two processes intersect to cause neuron death. If the two pathways intersect, which is upstream? A similar convergence of pathways related to mitophagy has been observed for autosomal recessive PD.

Approaches:

- Determine what is needed for disease-modifying therapy for autosomal recessive PD patients.
- Determine the best predictive models for preclinical evaluation.
- Explore phenotypically relevant mouse models of *SNCA* mutation crossed into *Park2* -/-, *Pink1* -/-, or mitochondrial *Polg* mutant (mutator) backgrounds or other mitochondrial-stressed mice to determine if there is synergy between α-synuclein aggregation and mitochondrial stress in causing dopaminergic neuron loss. One could also consider iPS cell models from patients to assess crossover between the two pathways *in vitro*.

<u>Recommendation 10</u>: Develop tools for measuring pathway architecture and flux in PD and integrate findings across analytical platforms into a systems level understanding of pathogenesis and a blueprint for effective therapeutic intervention.

Need: Many of the systems affected in PD and related diseases are dynamic, and involve protein and organelle trafficking. In contrast, the majority of studies do not examine the flux of molecules through these dynamic systems. Likewise, defects in (for example) mitochondrial homeostasis can also affect metabolism in different ways but how and where bottlenecks form in networks is largely unknown. One can think of PD mutations as promoting a change in state(s) of protein and metabolome networks. Moreover, protein modification states within crucial networks may also undergo a change in state. This involves both discovery-based quantification of networks and modifications in different cell states as well as targeted analysis of particular selected proteins and modifications.

Approaches:

- Determine what networks are most closely related to events that lead to disease and what technologies are best suited for elucidating changes in state and flux.
- Deploy established and emerging technologies that interrogate the dynamics of protein networks on a scale that was not possible even several years ago.
- Develop experimental cell systems, possibly including iPS cells, which properly query/model specific genetic defects linked with PD.
- Quantify protein modifications such as phosphorylation, ubiquitylation, and acetylation in both networks as well as at the global level.

BASIC RESEARCH RECOMMENDATIONS (11)

***Recommendation 1:* Develop transmission models of pathologic α-synuclein and tau, and determine the mechanisms of propagation, release, and uptake of misfolded α-synuclein and tau including the role of "strains."**

Need: Emerging evidence implicates cell-to-cell transmission of misfolded proteins through templated recruitment as a common mechanism for the onset and progression of several neurodegenerative disorders including α-synuclein and tau proteins in PD, PD dementia (PDD) and dementia with LBs (DLB). The "transmission hypothesis" for non-prion neurodegenerative diseases provides a plausible and compelling potential explanation for the stereotypical spread of pathological aggregates in PD/PDD, DLB, and other neurodegenerative diseases. Specifically, for α-synuclein and tau, aggregate-containing lysates and/or synthetic fibrils assembled from recombinant proteins template or seed their soluble counterparts to form fibrils in cultured cells and/or living animals, even without overexpression of the disease protein in PD models. Other evidence implicates distinct conformers or "strains" of misfolded α-synuclein and tau as the molecular basis for remarkable disease heterogeneity and co-morbidities. For example, one α-synuclein strain preferentially recruited monomeric tau to seed formation of neurorfibrillary tangles (NFTs), the signature lesions of AD. NFTs also are common in PDD since a third of patients with PDD show concomitant AD pathologic changes in addition to abundant cortical LBs. However, our knowledge related to the concepts of transmission and strains is rudimentary. Hence, there is an urgent need to develop an in-depth understanding of these processes, as well as to elucidate the mechanism(s) of disease protein spread, in order to identify novel targets for PD therapies.

Approaches:

- Establish and characterize transmission models of pathologic α-synuclein and tau in non-transgenic mice, rats, and non-human primates to model more authentically PD, PDD, DLB, and other synucleinopathies.
- Determine the sequence and structural determinants of α-synuclein and tau that are essential for cell-to-cell transmission and spreading of pathologic α-synuclein and tau, and the potential role of distinct α-synuclein and tau strains to cross-seed each other, as well as transmit different forms of PD, DLB, and PDD.
- Identify mechanisms for release of α-synuclein and tau pathological conformers that transmit disease, and for the uptake of these pathological conformers by other normal neurons and glial cells *in vitro* and *in vivo*.
- Elucidate mechanisms underlying the recruitment and corruption of endogenous normal α-synuclein and tau proteins to form fibrils, as well as the normal cellular processes that fail to block seeded propagation and accumulation of these pathologies in cell and animal models in PD and other synucleinopathies.
- Determine how heritable genetic factors (*e.g.*, mutations and polymorphisms in the genes encoding *GBA, LRRK2, MAPT, PARK2, PINK1, SNCA*) modulate transmission of α-synuclein and tau pathologies in genetically engineered mice.

<u>Recommendation 2:</u> Elucidate the normal and abnormal function of α-synuclein and its relationship to other PD genes (e.g., ATP13A2, GBA, LRRK2, PINK1, and PARK2).

Need: The amount of wild type α-synuclein protein expressed appears to be a strong predictor of risk for PD, in both familial and sporadic forms of the disease. Although it is generally considered that the amount of protein expressed influences the risk of misfolding and the acquisition of abnormal, pathologic function, we do not know whether an increase in the normal function of α-synuclein also contributes to degeneration. Aggregation may in fact lower the amount of soluble α-synuclein, reducing its function with pathologic consequences. It will also be important to understand the effects of reducing α-synuclein, since many current therapeutic approaches target the protein. In addition, changes in conformation associated with the normal function of α-synuclein may predispose it to misfolding or contribute directly to pathologic effects when misdirected to organelles with which α-synuclein does not normally associate, such as mitochondria and lysosomes. Neural activity may influence the behavior of α-synuclein, and conversely, α-synuclein may influence basal ganglia circuits. The apparent requirement for α-synuclein in 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) toxicity further implicates the normal function of the protein in degeneration. For all of these reasons, it is essential to elucidate the normal function of this protein, which will be important even if many current theories about pathogenesis are either not correct or not relevant. It will be particularly important to elucidate the relationship of α-synuclein to other PD genes since many cause characteristic α-synuclein accumulations.

Approaches:

- Use α-synuclein knockout mice to elucidate the normal function of the protein. Better cell-based assays will become important to distinguish the direct effects of α-synuclein from indirect effects due to changes in membrane lipid composition, for example, and to reassess the effect of PD-associated mutations. It will also be important to understand how the family of synuclein proteins, including β- and γ- as well as α-synuclein, influence transmitter metabolism and sensitivity to exogenous toxins.
- Determine how loss of α-synuclein function influences the behavior of basal ganglia circuits. α-Synuclein may have physiological effects that are not cell-autonomous and underlie its role in degeneration.
- Identify the physiological mechanisms that regulate α-synuclein expression *in vivo*. Considerable attention has been paid to clearance of the protein, but α-synuclein was independently identified on multiple occasions as a gene induced by a range of stimuli from growth factors to toxins.
- Use the information about normal function of α-synuclein to guide the analysis of knockout mice lacking GBA, LRRK2 and other PD genes and determine their epistatic relationship.
- Elucidate the relationship between α-synuclein and organelles implicated in PD, in particular mitochondria and lysosomes.
- Determine whether dysfunction precedes or follows protein aggregation in patients with PD. In addition to post-mortem analysis, this will require early identification of patients at risk by non-motor symptoms, genetics or imaging (which will in turn depend on a better understanding of the relationship between aggregation of recombinant α-synuclein *in vitro* and LBs).

<u>Recommendation 3:</u> Understand how different cell populations change in their coding properties, firing patterns, and neural circuit dynamics over time; how these changes relate to behavior and motor control; and how therapeutic interventions may affect such changes.

Need: Neuroscience is uncovering evidence for large-scale network activity dynamics and neuroplasticity in brain circuits, including those thought to be core circuits disabled in PD. This opens up the possibility of determining with precision how PD-related circuits encode information; become altered by experience; and react to loss of dopamine, changes in oscillatory patterning, and maladaptive anatomical and gene expression

changes in the parkinsonian state. We urgently need more information about these PD-related circuits, including as much information as possible about the specific cell types (neurons, glia) and molecules (neurotransmitters, neuromodulators, genes and epigenetic modifiers) involved in these pathways. There is an unprecedented opportunity, with the explosion of new methods coming into neuroscience and medicine, to fill the enormous gap between what we would need to know to design better therapeutics and what we actually know about PD-related circuits.

Approaches:

- Map circuits with state-of-the-art electrophysiological and anatomical methods to develop a circuit plan including connectivity, activity dynamics, and micro-circuit interactions.
- Apply circuit manipulations at key mapped nodes by advanced optogenetic and related (*e.g*., designer receptors exclusively activated by designer drugs) manipulations and microfluidic methods, as well as improved models of DBS.
- Apply chronic, not only acute, monitoring techniques of electrical activity in PD circuits combined with neurochemical activity monitoring, *e.g*., fast-scan voltammetry, Ca^{2+} imaging, and two-photon imaging.
- Develop next-generation neuro-feedback techniques for therapeutic use to reconfigure vulnerable circuits.

***Recommendation 4:* Generate and characterize a panel of PD-specific iPS cells (sporadic and genetic, including isogenic lines) for "omic" (RNA sequence, proteomics, methylation, etc.) pathway analysis and other approaches.**

Need: Recent developments in the reprogramming of human somatic cells to pluripotency with defined factors have the potential to revolutionize the study of the underlying pathogenesis of a variety of human disorders. PD is the most common movement disorder that is due, in part, to the preferential loss of dopamine neurons. The relative selective degeneration of dopamine neurons makes PD a particularly attractive human neurodegenerative disease to establish patient-specific cells in culture. Successful implementation has the potential to transform the study and treatment of PD by providing new molecular insights into the pathogenesis of PD. Moreover, the potential discovery of biochemical and/or molecular markers ultimately could be used as biomarkers to monitor the progression of PD.

Approaches:

- Characterize genetic and sporadic PD–specific iPS-derived neurons and glia at the cellular, physiologic, molecular, genomic, and proteomic levels.
- Develop facile and rapid methods for differentiating iPS cells into mature dopamine neurons and apply rigorous tools to interrogate differentiated human dopamine neurons, including cell-sorting markers.
- Develop and refine methods for studying the molecular and physiologic properties of transplanted human dopamine neurons in rodent and non-human primate models
- Use PD-specific iPS-derived neurons and glia to elucidate mechanisms of neurodegeneration that are relevant to PD and related disorders.

***Recommendation 5:* Integrate comprehensive datasets and perform functional and genetic analyses across large data sets.**

Need: There has recently been an explosion of information derived from large-scale experimental approaches in PD-focused research. These comprehensive analyses include, but are not limited to, genetic screens in model organisms, expression and epigenomic analyses in patient tissue and appropriate models (iPS cells, mouse and other mammalian models), GWAS, and large-scale drug screens. While all these approaches are

important individually, there now exists a critical unmet need to integrate comprehensive data sets to identify most effectively pathways and mechanisms that impact key disease phenotypes and pinpoint the most promising therapeutic targets.

Approaches:

- Gather and integrate data from multiple comprehensive data sets, including model organism and human genetic studies, expression, and epigenomic analyses in a variety of relevant model systems and comprehensive drug screens. Create a system to promote rapid and open data dissemination from all large-scale screens.
- Develop and implement novel and effective tools of analysis and integration of these large data sets. This would likely include a "genome browser"–style interface, and creation of a PD-specific pathway/network that can be browsed and augmented.
- Utilize data sets derived from patient populations, cell types, and models that can address pathways involved in both motor symptoms and key non-motor manifestations of PD to better understand non-motor symptoms in PD.
- Apply a more system-wide approach in many of these efforts to understand the interplay of chronological age, the genome, the epigenome, gene expression and splicing, and protein modification in the context of a cell system or vulnerable tissue to provide key mechanistic insights.
- Validate pathways and proteins identified through large-scale data set integration and system wide data analysis approaches mechanistically in the appropriate experimental models.

***Recommendation 6:* Develop approaches to exploit direct access to the human brain in individuals with PD during neurosurgical procedures such as DBS, for example using non-invasive high-resolution magnetic resonance imaging and positron emission tomography.**

Need: Although a number of animal models of PD have been developed and new models continue to advance, no model is yet able to recapitulate the spectrum of features, either symptoms or neuropathologic changes, or the time course of disease progression in humans with PD. Thus, it is critical to advance studies in human subjects, and several recent advances have made this possible. First, the rapid growth of functional neurosurgical procedures, especially DBS, provides unprecedented direct intraoperative access to the human brain. In parallel, continued development of non-invasive imaging modalities such as high-resolution MRI and PET – for example, the development of 7T MRI and high-resolution research tomography PET - provide approaches to quantify the structural and biochemical changes that occur during the onset and progression of PD. These reverse translation activities are essential parallel adjuncts to studies in animal models to understand, validate, and improve the relevance of these animal models for both basic science PD research and translational efforts.

Approaches:

- Develop and validate hardware and software tools that enable intraoperative perturbation and recording of relevant variables (electrical, neurochemical) in the human brain, including regulatory strategies to enable application and dissemination of these tools.
- Establish quantitative relationships between patterns of neural activity, including electrical, chemical, and metabolic, and symptoms to track disease progression and establish relevant biomarkers.
- Develop contrast agents/ligands to enable quantitative imaging of structure and biochemistry in the parkinsonian brain.
- Conduct parallel studies in humans and animals to understand better the limitations of current animal models to provide therapeutic leads.

<u>Recommendation 7:</u> Develop a more detailed understanding of the genetic basis of PD.

Need: Genetic work in PD has expanded from its initial success in monogenic forms of the disease to include understanding of the genetic architecture of apparently idiopathic PD. The field has now put forth a major collaborative effort in genetics, resulting in the identification of 28 independent loci for disease risk. While this progress is beginning to shed insight into the basic etiologic processes (*e.g.*, Rab-7L1 and LRRK2) more needs to be done. The immediate needs are threefold: 1) moving from our understanding that a locus is associated with disease to proving which transcript is the biological effector of this association, 2) expanding our genetic dissection of risk, and 3) extending the genetic efforts beyond simple risk, to include disease-related traits such as age at onset, progression, presentation of motor and non-motor features, and response to treatment. While each of these will shed light on the basis of disease, the last point is essential to understand disease heterogeneity and for precision medicine for PD. The majority of these projects require large collaborative efforts.

Approaches:

- Determine the detailed role of genetic risk variability on gene expression (including splicing) and protein expression (including isoforms and modifications) in relevant tissue, including iPS-derived neurons and glia. This would require a major effort assessing 100's of candidates in large numbers of samples.
- Resequence known PD loci in thousands of samples to identify common variant risk alleles, and rare disease-linked variants.
- Sequence exome/genomes in tens of thousands of samples. This will take the form of pooling extant data and investment in new sequencing.
- Assess in depth genotype/phenotype relationships, including comprehensive genetic analysis in cohorts with existing longitudinal data and development of new cohorts for longitudinal assessment. In the first instance genetic approaches are likely to be limited to evidence based candidate assessment, but can be expanded to genome wide discovery with sufficient numbers.

<u>Recommendation 8:</u> Develop a more detailed understanding of the molecular determinants and mechanisms of α-synuclein and tau aggregation (oligomer and fibril formation), disaggregation and clearance.

Need: Several lines of evidence demonstrate that α-synuclein aggregation plays a central role in the etiology of PD (both familial and sporadic forms) and that tau aggregation contributes to other neurodegenerative disorders characterized by parkinsonism. However, very little is known about the structure and dynamic properties of different α-synuclein and tau aggregates such as oligomers, fibrils and LBs and NFTs, and the molecular determinants, cellular mechanisms, and pathways that regulate their formation, clearance, and toxicity. In addition, it remains to be determined what role sequence variants and post-translational modifications play in modulating these processes. Several factors contribute to this gap in knowledge, including the lack of model systems that accurately recapitulate α-synuclein and tau aggregation and accumulation of LBs and NFTs in the brains patients with parkinsonism, lack of experimental approaches to observe directly these processes; and difficulties in isolating intact α-synuclein in LBs and tau in NFTs from human brains. Filling this knowledge gap by delineating α-synuclein and tau aggregation, clearance, and functional pathways will facilitate development of novel therapies for neurodegenerative disorders characterized by parkinsonism.

Approaches:

- Develop and apply state-of-the-art biophysical and high-resolution imaging techniques including single molecule microscopy to achieve both qualitative and quantitative characterization, as well as real time monitoring of structural properties to study the dynamics of α-synuclein and tau oligomerization, fibril

formation, dissociation, and clearance *in vitro,* in cells, in animal models, and in human biological samples.

- Determine the role of post-translational modifications in regulating α-synuclein and tau aggregation, disaggregation, and clearance *in vitro* as well as in cellular and *in vivo* models of neurodegenerative parkinsonian synucleinopathies and tauopathies.
- Develop and validate cellular, organotypic slice culture, and animal models that recapitulate more faithfully pathologic α-synuclein and tau and/or toxicity in the human brain. Emphasis should be on creating models that accurately recapitulate the location and expression levels of α-synuclein and tau *in vivo* to avoid potential artifacts associated with non-physiological contexts.
- Determine the role of the cellular environment, *e.g*., presence of dopamine, membrane interactions, neuroinflammation, in the oligomerization and fibrillization of α-synuclein and tau.
- Determine the mechanisms by which tau and tau aggregation influence the aggregation and toxicity of α-synuclein and vice versa.

<u>Recommendation 9:</u> Use a combination of sensor technologies and imaging to develop a more precise understanding of the neural circuit dynamics in PD to enable the development of next-generation therapeutic devices.

Need: Despite the established clinical efficacy, the mechanism of DBS in PD is incompletely understood. Because ablative neurosurgery for PD is similarly effective for treating PD, the stimulation-evoked silencing of pathologically hyperactive neurons was initially postulated as the primary mechanism. However, more recent studies have reported activation of output nuclei from DBS target structures such as subthalamic nuclei (STN) and globus pallidus interna (GPi). The neural network activation hypothesis has enormous implications for DBS mechanism of action. Indeed, DBS should evoke target-specific changes in neural activity in interconnected structures within the basal ganglia complex that ultimately underlie clinical benefit. Nevertheless, our understanding of these distal effects of DBS remains far from complete, in large part because of the technical difficulties in using imaging and sensor technologies for global assessment of neural activity in animal models and in human patients with an implanted device. Further refinement of our understanding of the mechanism of DBS is critical to enable the optimization of DBS for patients with PD.

Approaches:

- Use state of art sensor technologies (such as fast scan cyclic voltammetry or amperometry) to interrogate the dynamic neurotransmitter and related molecule changes that occur within the DBS target structure, as well as their functionally interconnected basal ganglia complex, and correlate with clinical outcome measures.
- Develop combination imaging and sensor technologies that can be deployed in human patients and animal models to allow for global network assessment during DBS.
- Develop predictive mechanistic models of neural circuit dynamic changes during DBS using precise target-specific activation patterns on imaging and neurotransmitter dynamic changes.
- Use understanding of mechanism of action of DBS to develop next-generation therapeutic devices that utilize closed loop architecture.

<u>Recommendation 10</u>: Develop a more comprehensive understanding of the role of catabolic pathways in PD, including assessment of both the ubiquitin-proteasome and the autophagy-lysosomal systems.

Need: Substantial experimental evidence suggests that disturbances in cellular catabolic pathways are central to the pathogenesis of PD. Human genetic studies, cell biology approaches, and animal studies have implicated disturbances in either or both of the two major intracellular catabolic pathways: the ubiquitin-

proteasome system and the autophagy-lysosomal system. In some cases (*e.g.*, *GBA* or *ATP13A2*) a global cellular disturbance in the catabolism of macromolecules is implicated. In other cases (*e.g.*, *PINK1* or *PARK2*) more discrete catabolic defects may be at work. Yet despite compelling evidence implicating catabolic defects in PD, substantial gaps in knowledge remain. Our knowledge of the normal role of PD-related genes in the functioning of catabolic pathways and how this functioning is impacted by disease variants is incomplete. The downstream consequences of catabolic pathway defects on cellular physiology relevant to PD remain to be fully elucidated. Finally, the pervasiveness of these putative defects in catabolic pathways among different subgroups of patients with PD is unclear. Interdisciplinary approaches that integrate human genetic studies with genetically tractable models, mammalian models, and emerging human iPS cell models are needed.

Approaches:

- Determine the normal role(s) of PD-associated genes in the functioning of the ubiquitin-proteasome system, the autophagy-lysosomal system, or cross talk between these systems, and how disease-associated variants alter these activities.
- Determine the downstream consequences of PD-related catabolic pathway defects on cellular physiology (*e.g.*, synaptic function, mitochondrial function).
- Assess the contribution of catabolic pathway defects to promoting or limiting other pathogenic mechanisms implicated in PD (*e.g.*, cell-to-cell spread of α-synuclein and/or tau).
- Elucidate the pervasiveness and severity of catabolic pathway defects in familial and sporadic populations of patients with PD (*e.g.*, early onset vs. late onset, sporadic vs. familial, association with specific genetic variants).

<u>Recommendation 11:</u> Advance our understanding of neural circuits, circuit analysis techniques, PD animal models, and optogenetic and related imaging technologies to improve existing therapies and generate next-generation therapies for PD.

Need: Treatments that ameliorate symptoms of PD modify the dynamics of large-scale neural circuits. Yet, due to the complexity of the neural circuits that go awry in PD, and our still-limited understanding of their normal and pathophysiological states, more effective treatments would undoubtedly result from a clearer understanding of the circuitry and its dynamics. Recently, a new generation of technologies has emerged that collectively hold the potential to answer many of the outstanding questions about the neural circuits involved in PD. These exciting technologies include optogenetic means for activating and inhibiting specific cell types using light, imaging techniques for visualizing the dynamics of genetically defined cell types during animal behavior, computational methods for analysis of large-scale imaging data, methods of high-resolution circuit reconstruction, and genetically encoded fluorescent sensors that report neural Ca^{2+} excitation, voltage depolarization, or release of neurotransmitter or neuromodulator. Application of these approaches in animal models of PD will likely yield substantial insights into circuit structure and dynamics, as well as improved therapies based directly on these insights.

Approaches:

- Identify, using imaging techniques that reveal the dynamics of genetically identified cell types in freely behaving animal models, normal patterns of neural circuit activity, how these patterns relate to mammalian behavior, and how these patterns and behavior go awry in PD.
- Create high-resolution maps of how PD alters both long-range and local neuronal micro-circuitry, and relate these anatomical and cytoarchitectural changes to the alterations in circuit dynamics and mammalian behavior. Ideally, these maps should be based on the same individual animals studied while alive in large-scale studies of neural dynamics and behavior, to facilitate detailed conceptual links between the various data sets.

- Develop suitable computational methods that can characterize and classify large-scale imaging data sets, in such a manner that these computational methods could facilitate next-generation, large-scale screens for new pharmacologic or DBS treatments using large colonies of animals and automated computational assessment of treatment efficacy.
- Use imaging techniques in freely behaving animals, in conjunction with fluorescent sensors of Ca^{2+}, voltage, and transmitter/modulator release dynamics, to evaluate in identified cell types the acute and long-term effects of existing treatments, either pharmacological or DBS, and to assay new therapies.
- Extend existing optogenetic and imaging techniques that are chiefly used today in rodent models of PD so that these technologies can be readily used in primate models of the disease.
- Develop tandem methodologies in PD animal models, combining measurements that can be performed in human subjects with optogenetic and imaging methods that are normally restricted to animals to identify novel disease biomarker. Successful outcomes will involve measurements that can be performed clinically as well as an understanding of what the novel biomarkers reflect at the microscopic or circuit-level scales.

Summary Tables:
NINDS PD2014 Research Recommendations

Table 1: Highest Priority Recommendations in Each Research Topic Area		
Topic Area	**Recommendation**	
Clinical Research	1	Define the features and natural history of prodromal PD including progression, events that underlie phenoconversion to clinically manifest PD, and biomarkers or other determinants of prodromal subtypes with the goal of providing sufficient rationale to initiate proof-of-concept prevention trials that initially target high-risk populations.
	2	Develop effective treatments and companion biomarkers for dopa-resistant features of PD. These features include both motor symptoms, particularly gait and balance problems, such as freezing of gait, and non-motor symptoms, especially cognitive impairment, psychosis, and dysautonomia.
	3	Characterize the long-term progression of PD and understand the mechanisms that underlie the heterogeneity in clinical presentation and rates of progression. Factors related to disease heterogeneity may include clusters of clinical features as well as biological factors such as genotype and biomarkers.
Translational Research	1	Develop patient stratification tools that define disease signatures of more homogeneous cohorts with emphasis on slow- vs. fast-progressing PD, prodromal PD, and non-motor symptoms.
	2	Develop novel and specific PET imaging agents and assays to measure α-synuclein burden, validated in both animal models and human tissue.
	3	Develop resources with greater power to predict efficacy and biomarker outcomes in clinical trials. These resources would include well-characterized replication sets of iPS cell lines from sporadic, dominant, and recessive PD cases.
Basic Research	1	Develop transmission models of pathologic α-synuclein and tau, and determine the mechanisms of propagation, release, and uptake of misfolded α-synuclein and tau including the role of "strains."
	2	Elucidate the normal and abnormal function of α-synuclein and its relationship to other PD genes (*e.g.*, *ATP13A2, GBA, LRRK2, PINK1*, and *PARK2*).
	3	Understand how different cell populations change in their coding properties, firing patterns, and neural circuit dynamics over time; how these changes relate to behavior and motor control; and how therapeutic interventions may affect such changes.

Abbreviations. GBA: glucocerebrosidase, iPS: induced pluripotent stem, LRRK: leucine-rich repeat kinase, PET: positron emission tomography, PINK: PTEN-induced putative kinase.

Table 2: Additional Priority Recommendations for Clinical Research	
4	Develop biomarkers of target engagement and proximal pharmacodynamic effects for use in early stage clinical trials.
5	Improve methods to assess long-term efficacy and potential for disease modification in clinical trials, including more efficient strategies for screening potential agent and trial design simulations to assess their performance for predicting long-term benefits.
6	Determine factors that facilitate public health interventions, including risk factor reduction and health services interventions for populations and individuals.
7	Use innovative outcome measures to evaluate motor and non-motor features, including patient- and clinician-reported outcomes that leverage emerging information technology (IT) opportunities, enhance sensitivity and specificity of measurement, and facilitate long-term follow-up of well-characterized cohorts.
8	Develop improved informatics capability to include investigation of how "big data" may contribute to a fuller understanding of PD, a central repository for PD trial data, a resource for trial design simulations.
9	Develop strategies to increase minority participation in research. These initiatives should include mechanisms to assess the effectiveness of these programs and could lead to the establishment of shared resources to facilitate minority recruitment in PD clinical trials.
10	Identify risk factors and pathogenic mechanisms of motor fluctuations and dyskinesias to identify novel targets for prevention and symptomatic therapy.

Table 3: Additional Priority Recommendations for Translational Research	
4	Develop an integrated PD knowledge base that includes data from genetic, biomarker, and clinical research, and from clinical trials with informatics support for integration of existing databases for PD and other chronic neurodegenerative diseases.
5	Establish consensus guidelines for preclinical therapeutic studies targeting α-synuclein to ensure appropriate use of existing models, improve replication of results across labs, and provide recommendations for future model development.
6	Develop intermediate markers of drug efficacy in early PD translational studies to support more efficient proof-of-concept studies.
7	Define the required attributes of targets emerging from basic science efforts that justify advancement into translational studies in PD.
8	Develop a thorough understanding of targets and pathways associated with pathogenesis and pathophysiologic mechanisms of PD with emphasis on those validated by human genetics and biology.
9	Investigate the relationship between converging pathways in PD, for example α-synuclein misfolding and mitochondrial function.
10	Develop tools for measuring pathway architecture and flux in PD and integrate findings across analytical platforms into a systems-level understanding of pathogenesis and a blueprint for effective therapeutic intervention.

Table 4: Additional Priority Recommendations for Basic Research	
4	Generate and characterize a panel of PD-specific iPS cells (sporadic and genetic, including isogenic lines) for "omic" (RNA sequence, proteomics, methylation, *etc*.) pathway analysis and other approaches.
5	Integrate comprehensive datasets and perform functional and genetic analyses across large datasets.
6	Develop approaches to exploit direct access to the human brain in individuals with PD during neurosurgical procedures such as DBS, for example using non-invasive high-resolution magnetic resonance imaging and positron emission tomography.
7	Develop a more detailed understanding of the genetic basis of PD.
8	Develop a more detailed understanding of the molecular determinants and mechanisms of α-synuclein and tau aggregation (oligomer and fibril formation), disaggregation and clearance.
9	Use a combination of sensor technologies and imaging to develop a more precise understanding of the neural circuit dynamics in PD to enable the development of next-generation therapeutic devices.
10	Develop more comprehensive understanding of the role of catabolic pathways in PD, including assessment of both the ubiquitin-proteasome and the autophagy-lysosomal systems.
11	Advance our understanding of neural circuits, circuit analysis techniques, PD animal models, and optogenetic and related imaging technologies to improve existing therapies and generate next-generation therapies for PD.