LRRK2 as Therapeutic Target

Jan Egebjerg
Scientific and Medical Rationale(s)

**Scientific**
- Strong genetic evidence causally associates LRRK2 to familial PD.
- Combined genetic and biochemical evidence supports a hypothesis in where the LRRK2 kinase function correlates with disease risk and that LRRK2 kinase inhibitors would be a new treatment paradigm for PD.
- Expression of LRRK2 is highly enriched in brain, lung, kidney and blood.
- Successful LRRK2 inhibition would successfully dampen LRRK2 kinase activity in the brain with a sufficient TI.

**Medical Rationale**
- Current PD treatments treat symptoms and have no effect on disease progression and limited of effect at late stages of disease.
- Large unmet need for effective treatments and in particular treatments that may alter the progression of the disease or even modifying the disease.
How difficult can it be?

- Identify a selective LRRK2 kinase inhibitor
- Target engagement: Tools to determine target engagement in brain
- Pharmacological relevance: Disease relevant model for dose finding and time for intervention
- Patient selection and outcome measures for clinical trials
LRRK2 as a target for some PD patients?

- **Disease hypothesis:** Increased LRRK2 kinase activity causal for Parkinsons Disease

- **Patient population?**
  - Target only G2019S carriers: develop a LRRK2;G2019 S selective compound
  - Develop a selective "pan-LRRK2" inhibitor and identify patients with increased risk or LRRK driven pathology
Highly potent and "selective" LRRK2 kinase inhibitors have been identified.

- Challenging target (no approved CNS active kinase inhibitors) – transition peripheral target properties to CNS drug properties
- High quality compounds have been shared with the community

**Patenting Activity 2013-2015**

<table>
<thead>
<tr>
<th>Company</th>
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<tbody>
<tr>
<td>Merck</td>
<td>Sanofi</td>
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<tr>
<td>Genentech</td>
<td>Southern Research Institute</td>
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<tr>
<td>Pfizer</td>
<td>Arrien Pharmaceuticals</td>
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<td>GSK</td>
<td>Elan</td>
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<tr>
<td>Lundbeck</td>
<td>Zenobia</td>
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<tr>
<td>Origenesis</td>
<td>Cellzome</td>
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<td>Ipsen</td>
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- Issue: PK properties for appropriate therapeutic index (TI)
- Pharmacodynamics: Dose finding based on a pathophysiological relevant readout
**Tg rodent models**

**Leucine-Rich Repeat Kinase 2 Regulates the Progression of Neuropathology Induced by Parkinson’s-Disease-Related Mutant α-synuclein**

Xian Lin,¹,² Louka Paraskeacou,¹,² Xing-Long Gu,¹,³ Lizhen Wang,¹,³ Moon Shim,¹,³ Lin Xu,¹ Chengsong Xie,¹ Caixia Long,¹ Wan-Jou Yang,¹ Jinru Ding,¹ Zhuo-Zhu Chen,¹ Paul E. Gallant,¹ Jung Hwa Tso-Cheng,¹ Gay Rudow,¹ Juan C. Troncoso,¹ Zhiliu Li,¹ Zheng Li¹ and Huibin Cai¹

**Enhanced Striatal Dopamine Transmission and Motor Performance with LRRK2 Overexpression in Mice Is Eliminated by Familial Parkinson’s Disease Mutation G2019S**

Xiating Li¹,²,³ Jirot C. Patel¹,² Jing Wang¹,²,³ Marat Y. Avvalkamov⁴,⁵ Charles Nicholson⁶,⁷,³ Joseph D. Burcham,⁸,⁷,³ Gregory A. Elder⁶,³,⁵ Margaret E. Rice⁶,³ and Zhengyu Yue⁴,⁵

**R1441C mutation in LRRK2 impairs dopaminergic neurotransmission in mice**

Inhibitors of leucine-rich repeat kinase-2 protect against models of Parkinson’s disease

Byoung Dae Lee¹,²,³ Jooh-Shin Hwang¹,²,³, Jackalina VanKampen¹,²,³, Leonard Petrucelli¹,²,³, Andrew B West¹,²,³, Han Seok Ko¹,²,³, Yun-II Lee¹,²,³, Kathleen A Maguire-Zeiss¹,²,³, William J Bowers¹,²,³, Howard J Federoff¹,²,³,⁷,⁸ Valina I. Dawson¹,²,³,⁷ and Ted M Dawson¹,²,³,⁷,⁸

**The I2020T Leucine-rich repeat kinase 2 transgenic mouse exhibits impaired locomotive ability accompanied by dopaminergic neuron abnormalities**

Tatsunori Maejima¹,²,³,⁷,⁸ Sayuri Mori¹,²,³ Yui Sasaki¹,²,³,⁷ Takashi Miyajima¹,²,³,⁷ Sadahiro Azuma¹,²,³,⁷ Etsuro Ohta¹,²,³,⁷ and

**LRRK2 overexpression alters glutamatergic presynaptic plasticity, striatal dopamine tone, postsynaptic signal transduction, motor activity and memory**

Dayne A. Beccano-Kelly¹,²,³,⁷, Mattia Volta¹,²,³,⁷, Lise N. Muniesa¹,²,³, Sarah A. Paschall¹,²,³, Igor Tatarnikov¹,²,³, Kimberley Co¹,²,³, Patrick Chou¹,²,³, Li-Ping Cao¹,²,³, Sabrina Bergeron¹,²,³, Emma Mitchell¹,²,³, Heather Han¹,²,³, Heather L. Melrose¹,²,³, Lucia Tapia¹,²,³, Lynn A. Raymond¹,²,³, Matthew J. Farrer¹,³,⁷,⁸, and Austen J. Milnerwood¹,²,³,⁷,⁸

**Mutant LRRK²R1441G BAC transgenic mice recapitulate cardinal features of Parkinson’s disease**

Yanning Li¹,², Yencheng Liu¹,², Tianmiao F. Oo², Lei Wang³,⁴ Yi Tang⁴,⁵, Veronica Jackson-Lewis⁴,⁵, Chun Zhou⁴,⁵, Kindiya Geggma⁴,⁵, Milhaid Bogdanov⁴,⁵, Sergei Przedborski⁴,⁵, M Flint Beal⁶,²,³, Robert E Burke⁷ and Chenjian Li⁴

**Dopaminergic Neuronal Loss, Reduced Neurite Complexity and Autophagic Abnormalities in Transgenic Mice Expressing G2019S Mutant LRRK2**

David Ramonet⁷, Joao Paulo L. Daher²,³,⁴,⁵, Brian M. Lin²,³, Kloodjan Stafa²,³, Jaekwang Kim⁴,⁵, Rebecca Bamejje⁶, Marie Westerlund⁷, Olga Pietnikova⁴, Lilliane Glauser⁴, Lichuan Yang⁵, Ying Liu⁵, Deborah A. Swing⁶, M. Flint Beal², Juan C. Troncoso²,³, Michael McGeer⁴, Nancy A. Jenkins⁶,³, Neil G. Copeland⁷,¹, Dagmar Glatzer²,³, Bobby Thomas⁴, Michael K. Lee⁶,³, Ted M. Dawson²,³,⁷,⁸, Valina L. Dawson²,³,⁷,⁸, Darren J. Moore⁷,³,⁸

**G2019S LRRK2 activates MKK4-JNK pathway and causes degeneration of SN dopaminergic neurons in a transgenic mouse model of PD**

C-Y Chen¹,², Y-H Wang²,³, K-Y Chen²,³, K-J Lin²,³, T-H Yeh²,³, Y-P Cheng²,³, C-S Lu² and H-L Wang²
Dose finding based on a pathophysiological relevant readout: PD

- There is no validated preclinical *in vivo* model for disease progression in Parkinson’s Disease
- Rodent animals models that carry G2019S or other pathogenic variants do not present with Parkinson’s Disease i.e. a-syn aggregates
- Several tg models exhibit changes in locomotor activity and striatal dopaminergic tone
- Robustness of models an issue for drug testing

Sloan et al
Basal ganglia circuitry in a PD-like state:
STN burst firing and behaviour

Rat aSyn AAV model

Cortex

Striatum

Thal

Cylinder test

Coefficient of variation of the interspike interval (CV ISI).

Single unit recording
Findings – α-synuclein AAV rat model supportive of LRRK2 interaction- but still (several) inconsistencies

★ LRRK2 KO studies – Long Evans rat
  • LRRK2 KO modulates α-synuclein mediated burst firing
  • Effect on aSyn-pS129
  • No significant effect on behavior although a partial reversal has been observed

★ LRRK2 inhibitor studies – Sprague Dawley rat
  • Acute LRRK2 inhibition modulates α-synuclein mediated burst firing phenotype
  • Chronic LRRK2 inhibition modulates α-synuclein mediated behavioral phenotype
  • No significant chronic effect of LRRK2 inhibition on ephys
  • No significant effects on aSyn-pS129 after acute/chronic dosing
  • No significant effect on behavior after acute/chronic dosing
PK/PD modeling

PK/PD based on mechanistic readout

- Auto-phosphorylation correlates with occupancy and disease risk
- Direct P-1292 (not measurable in vivo)
- Indirect P-935 (correlates with occupancy) – PD marker for PK/PD modeling?
- Rab phosphorylation as measure for pathophysiological pathway (link to α-synuclein)

Dose qualification based on in vitro

- Hypothesis: Revert kinase activity of LRRK2 G2019S to the level of the protective form: IC80
Target engagement: Translational tools to determine target engagement in human brain

- Markers for human studies – mandatory!
- **PET ligands** - very challenging target – low abundance protein, lipophilicity of high affinity compound compromise signal/noise
- P-LRRK2 levels in CSF exosomes (or brain specific exosomes isolated from blood)
Clinical progression of Parkinson’s Disease (PD)

Topics to resolve in preclinical models:
Time for intervention: Is LRRK2 dysfunction critical at particular stages of the disease?
What readout would be most sensitive to LRRK2 kinase inhibition?
Model for clinical trial based on PPMI data (idiopathic PD patients)

- Early idiopathic PD (Hoehn & Yahr 2 or less)
- Positive DAT SPECT
- Time from diagnosis < 18mts
- +/- different concomitant treatments

- $\Delta$-DAT – scan
- $\Delta$-UDPRS
Modeling of change in disease progression

– Primary outcome at 1y: DAT imaging (change in striatum)
– Primary outcome at 2y: ∆UPDRS

<table>
<thead>
<tr>
<th>Reduction (% mean striatum)</th>
<th>Mean change (SD)</th>
<th>Total sample size 80% power</th>
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<tbody>
<tr>
<td>30%</td>
<td>-0.17 -&gt; -0.119 (0.20)</td>
<td>482</td>
</tr>
<tr>
<td>50%</td>
<td>-0.17 -&gt; -0.085 (0.18*)</td>
<td>144</td>
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<table>
<thead>
<tr>
<th>Difference (in Total Score)</th>
<th>Mean change (SD)</th>
<th>Total sample size 80% power</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 points (30%)</td>
<td>6.81 -&gt; 4.81 (12.07)</td>
<td>1146</td>
</tr>
<tr>
<td>3.4 points (50%)</td>
<td>6.81 -&gt; 3.4 (10.86*)</td>
<td>322</td>
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Segmentation strategies

- Exonic variants stratification
- **LRRK2 G2019S**
- + LRRK2 risk variant
- + PD minus LRRK protection carriers
- All PD patients

- Biomarkers for elevated LRRK2 activity
  - PBMC – phosphorylation state
  - Exosomes in urine
  - Others

- Symptom differentiators
**Focus area**

Increased focus on disease stratification

- Target/pathway specific markers for patient selection
- Biomarker approaches aiming at classifying patient heterogeneity

Use iPSCs as translational tool heterogeneity

- Patient disease pheno-/genotype defined iPSCs
  - Model disease heterogeneity
  - Confidence in target for dose estimation
  - Target identification

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Change from Baseline in UPDRS Scores by Patient (OFF State)
Progression/non-progression population defined at 2y and 3y post-baseline

**PATNO=3113**

**Progression**

**3113**

Change criteria: Change in MS>15
Non-change criteria: Change in MS<1

Change in UPDRS Motor Score (OFF state)

Change in UPDRS I-III Score (OFF state)

PATNO=3808

**Non-progression**

**3808**

Change criteria: Change in MS>15
Non-change criteria: Change in MS<1

Change in UPDRS Motor Score (OFF state)

Change in UPDRS I-III Score (OFF state)
How difficult is it?

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