**Meeting of the Minds | 思想的盛会**

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Most scientific meetings strive to inform the audience on the state of the field by presenting data in a consolidated manner, maximizing speaker time often at the expense of discussion time. Separating clinical and scientific sessions is common. This year, when planning our Investigator Meeting in Boston on May 21 – 24, Monica and Randy had the foresight to take a different approach.

在大多数的科学会议中，主办方都会力求让听众了解本领域的状况。因此会议总是按照一种习以为常的形式，牺牲掉讨论时间，从而把更多的时间给演讲者来呈现数据。此外，临床研究和科学研究也常常是各自以分会场的形式进行。但是今年，当我们筹划 5 月 21-24 日在波士顿召开的研究者会议时，Monica 和 Randy 有远见地采取了一种不同的方式。

They decided what we really needed was a “Meeting of the Minds” where our basic scientists and expert Rett clinicians could update one another on the current state of the field while keeping the focus on the key questions remaining to be addressed and the main obstacles to success. This potentially risky and unusual format focusing on challenges and difficulties, designed to foster active discussion and encourage brain-storming and problem-solving from new angles, was not only energizing and provocative, but effective. New approaches, out of the box thinking, and the seeds of new collaborations were sewn.

他们认为，所有人真正需要的是一场“思想的盛会”。这场盛会中，我们的基础科学家和 Rett 临床专家可以相互通达当前领域的研究状况，同时将注意力集中在有待解决的关键问题和通向成功的主要障碍上。这种将注意力集中在挑战和困难的反常形式会有一些潜在风险，但是能够促进积极的讨论，鼓励从新的角度进行头脑风暴和解决问题。从结果看，这种形式不仅充满活力和令人兴奋，而且有效。我们看到了新方法，跳出了固有思维，也孕育出了各种新的合作的种子。

The social aspects of the meeting helped to strengthen existing relationships and foster new ones. The investigators appreciated the opportunity to keep an eye on the ultimate goal by meeting several local children with Rett and learning about their lives. It was heartwarming to see the families acknowledge and appreciate the efforts of the Investigators to get to the bottom of this disorder.

会议的社交性部分体现在加强现有关系和培育新关系。研究人员很感谢有机会与当地的几个 Rett 孩子会面，了解她们的生活情况，以此来明确最终的目标。这些 Rett 家庭也在向研究人员表达感谢，感谢他们为彻底查明这一疾病的方方面面所做的努力，这也让研究人员倍感暖心。

The meeting started with the basic function of MECP2 and gene therapy approaches in development, followed by key tools and model systems in studying MECP2, MECP2 reactivation strategies and progress, and concluded with clinical research learnings and outcome measures to assess therapeutics.

会议以 MECP2 的基本功能和正在研发中的基因疗法开始，接着是研究 MECP2 的关键工具和模型，MECP2 再激活策略和进展，最后以讨论临床研究的发现和评价治疗方法效果的指标度量结束。

I came out of this meeting feeling extremely excited and optimistic about where things are heading, but also aware that challenges still lie ahead. In summary, the mutations that cause Rett Syndrome broadly impair brain function through complex mechanisms that are not completely understood. It is not surprising that addressing the root cause of Rett Syndrome by restoring MeCP2 protein in brain cells is profoundly more effective than therapeutics that target disruptions in one of the numerous downstream pathways. Because the mouse and human MECP2 genes are not identical, it will be necessary to optimize many of the newer biologic therapeutics (e.g., RNA modifications, MECP2 reactivation) in the human cell lines with stable X-inactivation that are currently being generated. While there is optimism that initial gene therapy will provide significant benefit, additional next generation gene therapy programs will continue to be pursued in parallel. A shared goal for these cure-focused programs is to obtain uniform delivery to all brain cells and effectively optimize MeCP2 protein levels in each cell.

当我从这次会议中走出时，我对将要发生的事情感到非常兴奋和乐观，同时也意识到前方依然有很多挑战。总体而言，导致 Rett 综合征的基因突变会通过一套复杂的机制在很大范围内损害了大脑的功能，而这套机制目前尚不完全清楚。因此，通过在脑细胞中恢复 MeCP2 蛋白质来除掉 Rett 综合征的病根，肯定远比在那些一大堆下游信号通路上的某一条上治疗更有效，这并不奇怪。此外，由于小鼠和人类的 MECP2 基因并不相同，因此也有必要对目前那些正在研究中的针对稳定的 X 染色体失活的人类细胞系的更新的生物疗法(例如，RNA 修饰，MECP2 再激活)进行优化。虽然人们乐观地认为，第一代的基因治疗就将带来明显的效果，但下一代的基因治疗项目也还将继续同步进行。这些项目的共同目标是能够找到一种方式对所有脑细胞统一递送药物的方法，并有效地优化每个细胞中的 MeCP2 蛋白质水平。

For anyone who wants to delve further into the research, below is more detail on what was presented in each session and some of the outstanding questions that will need to be addressed.

如果有人想要深入了解这些研究，那么请往下看，下面列出了各个分会场上讨论的详细内容，以及一些有待解决的关键问题。

**MECP2 Function**

**MECP2 的功能**

We know MECP2 regulates the expression of hundreds of genes and that behavioral changes in mouse models of Rett are difficult to assess in a meaningful way when evaluating potential therapeutics. The severity of MECP2 mutations in humans correlates well with the survival time of male Rett mice, suggesting that mouse survival may be a good marker for the potential success of therapies.

我们知道 MECP2 调控了上百个基因的表达，这种复杂性导致当用 Rett 小鼠模型来评估各种潜在的治疗方法时，很难找到一个好方法来评估这些小鼠在治疗前后的各种行为的变化。研究人员发现，对于 MECP2 基因的各种突变，其在人类中导致症状的严重度和雄性 Rett 基因突变小鼠模型的生存时间存在良好的相关性，由此表明小鼠的生

存时间可能会是某种有效果的治疗方式的良好标志。

A lively discussion ensued regarding the minimum level of MeCP2 protein required to improve cellular function, and what percentage of cells must attain this level to provide a therapeutic benefit. It was noted that survival in the mouse Rett model is meaningfully prolonged when MeCP2 protein levels are greater than 5-10% of normal in all cells or when normal levels of protein are achieved in 10-40% of cells.

接着，各位研究人员展开了关于 MeCP2 蛋白质的热烈讨论，讨论改善细胞功能所需 MeCP2 蛋白质的最低水平，以及有多少细胞达到这个水平才能表现出治疗的效果。研究人员发现，用小鼠模型评估时，当所有细胞总和的 MeCP2 蛋白水平高于处理前水平的 5-10%时，或者当 10-40%的细胞中 MeCP2 蛋白质达到正常水平时，Rett 小鼠的生存时间有显著延长。

**Genetic Approaches: DNA and RNA**

**遗传途径:DNA 和 RNA**

The most thought-provoking session at the meeting was gene therapy. A lively discussion acknowledged that gene therapy is a new therapeutic approach and that we are all pioneers. AveXis' Brian Kaspar indicated that all of the data points to a well-tolerated, non-toxic, gene therapy product, and presented efficacy results for AVXS-201 (heading into clinical trials early next year) that are profoundly better than any drug previously tested in mice.

基因治疗分会是本次会议中最引发众人思考的环节。研究者们进行了一场热烈的讨论，表明基因疗法作为一种新的治疗途径已经被认可，而且我们都是这个领域的先驱者。AveXis 的 Brian Kaspar 展示了 AVXS-201(目标明年初进入临床试验阶段)的疗效，所有的数据都表明这是一个有良好耐受性、没有毒性的基因药物，疗效比以前在小鼠身上试验过的任何药物都要好得多。

Subsequent presentations highlighted the risks associated with gene therapy as well as those unique for Rett Syndrome. However, we have confidence that FDA has sufficient experience with the gene therapy field to assure the study is as safe as possible for study subjects. Key unknowns will remain until the first clinical trial: will the MECP2 gene be delivered to a sufficient number of brain cells, and will the delivered gene produce an optimal amount of protein in each cell? We will not be able to reliably predict the magnitude of efficacy expected in this first trial. With this in mind, we will continue research in parallel focused on enhancing delivery to the brain and regulating the amount of MeCP2 protein produced.

随后的演讲则强调了基因治疗相关的风险，以及对 Rett 综合征来说特有的风险。不过，我们对 FDA 还是有信心的，相信 FDA 已经在基因治疗领域有充分的经验来保证临床研究时受试者尽可能安全。当然，在第一次临床试验之前，还有一些关键的未知因素：MECP2 基因会被送进足够数量的脑细胞吗？送进的新基因是否能够在每个脑细胞中生成理想数量的蛋白质吗？我们还没有办法去有效地估计在第一次临床试验中预期疗效的大小。考虑到这一点，我们还会继续同步研究，重点在于如何提高对脑细胞的基因输送效率以及更好调节 MeCP2 蛋白质的产量。

In a new and up-coming area of research, RNA modification, scientists are working to hijack existing cellular machinery to correct mutations at the level of RNA and thereby restore normal MeCP2 protein levels in each cell. Although considerably less advanced than gene therapy, these approaches offer advantages for regulating the level of MeCP2 protein in each cell. The RNA editing approach being pursued by the Mandel lab demonstrated the highest correction rate reported to date at a whopping 72%. RNA transplicing is an alternative approach that has the potential to correct over 97% of all MECP2 mutations with a single therapeutic. Current efforts are focused on optimizing the efficiency for modifying RNA and rapidly advancing development of these therapeutics toward clinical trials.

在 RNA 修饰这一新兴的研究领域，科学家们正试图接管现有的细胞工作机制，从而在 RNA 这一层面修正基因突变，进而恢复每个细胞中正常的 MeCP2 蛋白水平。尽管当前这些方法的进展并不如基因疗法，但是这些方法的优势在于能够调节每一个细胞中 MeCP2 蛋白质的水平。Mandel 实验室展示的一种目前正在研究的 RNA 编辑方法，其最高修正率高达 72%。RNA 剪切也是是一种替代方法，其有可能通过一种治疗方法纠正超过 97%的 MECP2 突变类型。当前工作集中在优化修改 RNA 的效率和将这些疗法向临床试验环节快速推进。

**Induced Pluripotent Stem Cells**

**诱导多能干细胞**

A key tool for developing many of the newer Rett therapeutics will be human induced pluripotent stem cells, hiPSCs. hiPSCs can be used to generate any other type of cell, allowing scientists to generate brain cells from Rett Syndrome patients. Scientists from academia and industry reported on progress in utilizing hiPSCs to discover and develop novel therapeutics for Rett Syndrome. Another tool being used to study Rett Syndrome is to generate organoids, or a “brain in a dish”, from hiPSCs. Organoids grow into more complex cell types and layers and provide informative ways to assess cell signaling.

人类诱导多能干细胞（human induced pluripotent stem cells，hiPSCs）将会是开发更多种新型 Rett 综合征治疗方法的关键工具。hiPSCs 可以被用来生成任何其他类型的细胞，这让科学家们能够得到 Rett 综合征患者的脑细胞。来自学术界和产业界的科学家们报告了利用 hiPSCs 发现和开发治疗 Rett 综合征的新疗法的进展。另一个可用来研究 Rett 综合征的工具是从 hiPSCs 中产生的类器官，通俗的说是“盘子里的大脑”。类器官可以生长成更复杂的细胞类型和层次，并提供了评估细胞信号传导的实用途径。

A unique issue in studying Rett Syndrome hiPSCs is that X inactivation, or silencing of one of the two X chromosomes in female cells, can degrade in these cell lines and we still don’t know how to fully control this consistently. It is important for the field to understand and control X-inactivation in hiPSCs not only to facilitate therapeutic development, but to explore the potential for exploiting underlying mechanisms to reactivate MECP2 on the silent X chromosome.

对于 Rett 综合征，在进行 hiPSCs 研究时有一个独特的问题，那就是在这些细胞系中 X 染色体的失活问题，换句话说在女性细胞中两条 X 染色体中的一个会沉默并在这些细胞系中退化。我们目前仍然不知道如何有效地控制 X 染色体失活。对 hiPSCs 研究来说，理解和控制 X 染色体失活不仅有利于研究治疗方法，而且还有可能探索挖掘出新的潜在方法来重新激活沉默的 X 染色体上的 MECP2 基因。

**MECP2 Reactivation**

**MECP2 再激活**

A key regulator of X inactivation is Xist, a non-coding RNA that coats the chromosome leading to its silencing. But is loss of Xist enough to turn on gene expression or do we need an additional activation step? Loss of Xist appears to have no negative effects in mice and can result in 2-5% reactivation of MECP2. A 4-10% increase in MeCP2 protein levels provides a 25% increase in mouse survival.

对于 X 染色体失活，一个关键的调控因子是 Xist，这是一种非编码的 RNA，它能够覆盖在染色体上从而使其沉默。但是，是否去除 Xist 就足以激活基因表达，还是我们需要一个额外的激活步骤？在小鼠模型上，去除 Xist 看上去似乎没有什么负面影响，并可让 2-5%的 MECP2 重新激活。这让 MeCP2 蛋白质水平增加了 4-10%，小鼠存活率增加了 25%。

Alternative approaches to activate MECP2 employing CRISPR/dCas9 or Zinc Finger guides are also being pursued. Recent experiments have shown that changing the state of the gene from a silenced signature to an active signature is enough to turn a gene on from the active X chromosome and conditions are being optimized to turn on MECP2 from the silenced chromosome.

另一种再激活 MECP2 的方法是用 CRISPR/dCas9 或锌指方法。最近的实验表明，将基因的状态从沉默的特征更改为活跃的特征就可以激活 X 染色体上的一个基因。研究者正在优化条件来特定激活沉默的 X 染色体上的 MECP2 基因。

Some key questions that remain are: Can this method be used in neurons and are the effects long or short-term? Will there be an immune response to the novel proteins administered to activate the gene? Can we use existing drugs to enhance the efficiency of MECP2 expression?

剩下的关键问题包括：这种方法是否可以用于神经元？其效果是长期的还是短期的？激活基因所需的外源蛋白质是否会产生免疫反应？我们可以用现有的药物来提高 MECP2 表达的效率吗?

**Clinical Research**

**临床研究**

The Natural History Study has enrolled almost 2,000 patients to date and the findings from this observational study have resulted in 30 publications with several pending or in preparation. Current efforts are focused on making these data more accessible to families, supporting industry and clinical trials in Rett Syndrome, and developing improved clinical outcome measures. Additional goals include standardizing clinical care and identifying biomarkers.

到目前为止，自然病史研究已经招募了近 2000 名患者。这项观察研究中发现的各种成果已经发表了 30 篇论文，还有几篇尚未发表或正在准备中。目前的工作重点是使让这些数据更容易被每个 Rett 家庭所了解，支持产业界和临床的 Rett 综合征试验，以及研究更好的临床结果测量指标。其他目标还包括临床护理方法的标准化和确定生物标记物。

Neurodevelopmental disorders are historically difficult to study and many successful therapies in mouse models fail to translate to efficacy in humans. It was suggested that paradigm shifts at FDA may be required with a focus to study the whole disease rather than specific behaviors. Physicians, advocacy groups, and industry are urged to engage FDA to develop improved assessments for efficacy in clinical trials. Can assessments in human cells improve the likelihood of efficacy in clinical trials for Rett Syndrome?

神经发育障碍历来都是很难研究的，许多在小鼠模型中成功的治疗方法转化到在人类身上时都未能体现疗效。有建议说 FDA 需要进行范式转变，将重点放在整个整个疾病的研究而不是一些特定的行为。这需要有医生、宣传团体和产业界一起和 FDA 合作，改进在临床试验中对疗效的评估方法。比如，在 Rett 综合征的临床试验中，能否通过对人体细胞进行评估来提高临床疗效成功的可能性？

**Novel Outcome Measures**

**新的结果测量指标**

Measuring change in disease symptomology is key to finding and demonstrating successful therapies. Rett specific severity scales that are better able to document meaningful improvements for individuals with Rett Syndrome and their families are clearly needed. A number of physiologic measures (brain waves, breathing patterns) are altered in Rett patients and could become objective biomarkers or outcome measures.

找到和确认某种疾病的成功治疗方法的关键是能够对这种疾病症状严重程度建立起可度量的指标。当前显然需要一个更好的 Rett 病情严重程度量表以便 Rett 综合征患者和他们的家庭记录下各种有意义的改善。此外，在 Rett 患者身上有许多生理指标（脑电波，呼吸模式）也和常人不同，这些也可能作为客观的生物标志物或测量指标。

In addition to typical outcome measures performed during a clinic visit, obtaining unbiased, objective data of a patient's status over a continuous period of time is emerging as an area of active development. The potential to employ biosensors and video recordings in the home environment provides an opportunity for continuous measurement of health, function and quality of life in a real world setting. Additionally, cellular protein and metabolic signatures are being explored to define disrupted cellular networks and potential corrective therapeutics, and to identify biomarkers for use in clinical trials.

在临床随访期间进行的常规指标的测量之外，连续长时间获取患者的公正客观的数据也渐渐成为一个积极发展的领域。技术的发展使得可以在家庭环境中有机会使用生物传感器和视频记录等方式来为现实环境中持续测量健康、能力和生活质量。此外，细胞中的蛋白质和代谢特征也在被考虑用于确定细胞网络的破坏程度和评价潜在的治疗方法，并作为临床试验中考量的生物标志物。