
Advancing Science
Zynerba®
PHARMACEUTICALS
Improving Connections
Recognizes
Fragile X Awareness Day

Zygel™ (ZYN002) Development Program in Fragile X Syndrome

17th NFXF International Fragile X Conference- Virtual series

July 22, 2020 Fragile X Research Roundup

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Zynerba Pharmaceuticals,

Chief Medical Officer

Disclaimers

- This presentation is intended to communicate scientific and medical information about data from clinical trials involving ZYN002. ZYN002 is an experimental treatment which has not been approved by any government regulatory bodies, including the United States Food and Drug Administration (FDA) and has not been determined by the FDA to be a safe or effective treatment for any disease or condition. This presentation is not designed or intended to promote the use of a ZYN002 or any other Company product in order to impact prescribing.

- This slide presentation is based on 3 abstracts submitted and accepted for presentation at the National Fragile X Foundation (NFXF) annual conference, originally scheduled for July 16-19, 2020. The meeting was subsequently cancelled due to COVID 19; alternatively, this virtual meeting was offered by NFXF to share abstract related presentations.

- Joseph Palumbo is a full-time employee of Zynerba Pharmaceuticals, Inc.

CONNECT-FX: Commitment, Dedication & Achievements

Families, Participants, Advocacy Groups, and Study Sites

NFXF

~1412 Patient Visits

8 Time zones

>4300 Caregiver Assessments + Skin Diaries

3 Countries

>63 Remote Visits

FRAXA

11 NFXF Center Sites

300 Children & Adolescents Screened

245 Participants Enrolled

Fragile X Association of Australia

>60 Site Staff

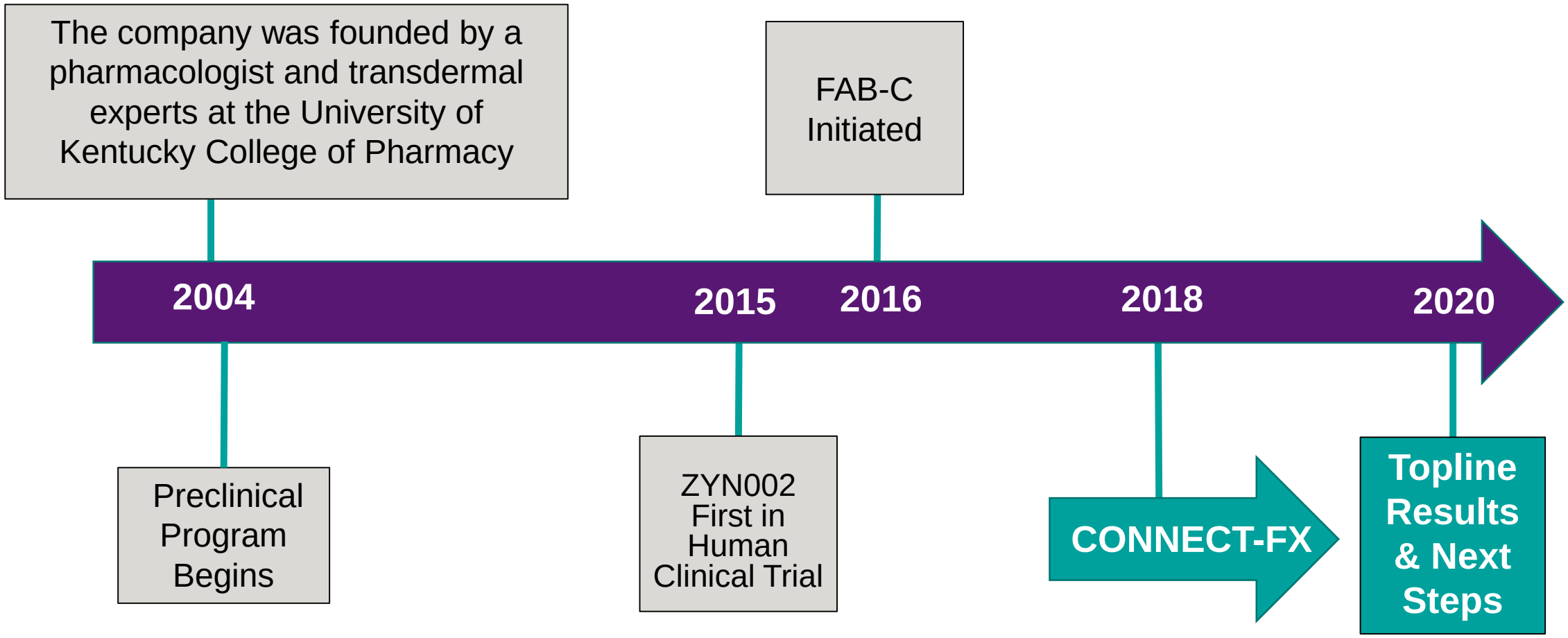
Travel by Planes, Trains, & Automobiles

~717 Blood Draws

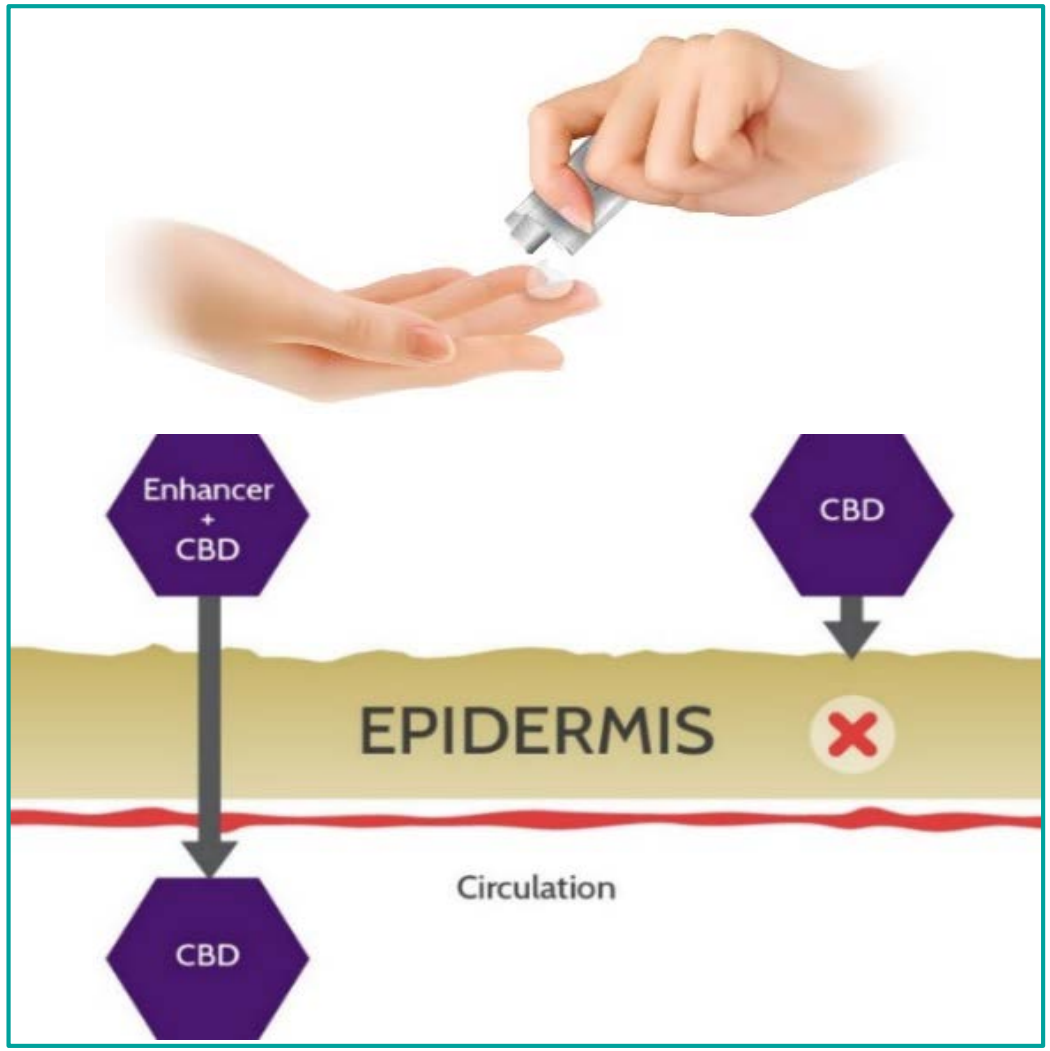
28 Person Zynerba Team

The History of Zygel (ZYN002) in FXS

More Than 15 Years of Research, Dedication, and Expertise



Key Characteristics of ZYN002



Transdermal (non-oral).
Avoids first pass metabolism.
Associated with few gastrointestinal adverse events across our clinical development program.*

Pharmaceutically manufactured.

Known purity and consistency.

No potential for pesticides or heavy metals; regulated manufacturing.

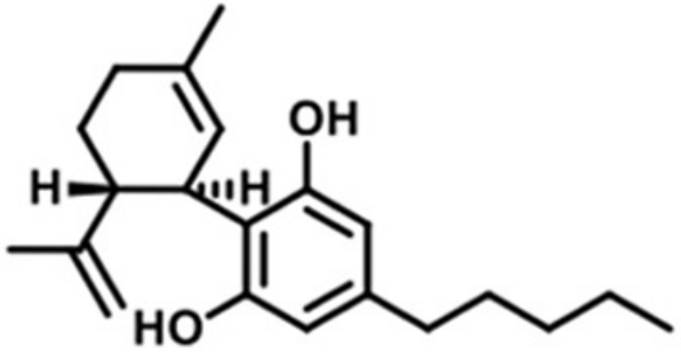
THC not detected in urine or plasma.*
Supports the purity and stability of CBD in ZYN002.

*Data on file.

Cannabidiol and Tetrahydrocannabinol

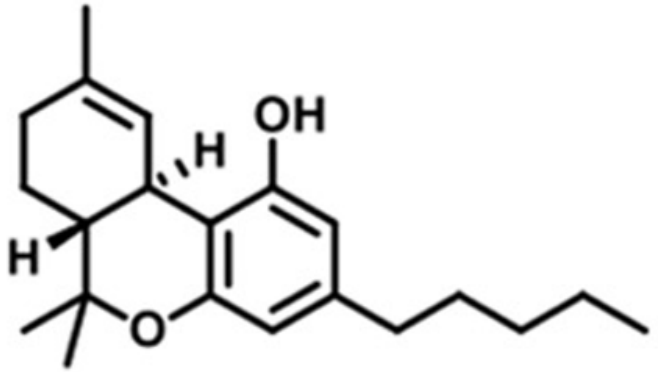
Cannabidiol (CBD)[1]

CBD is the non-euphoric component of cannabis



Tetrahydrocannabinol (THC)[1]

THC is the component of cannabis associated with euphoria



1. Elmes MW, et al. *J Biol Chem.* 2015;290(14):8711-8721.

The Endocannabinoid System (ECS) and FXS

Background

- The ECS consists of receptors in the brain and peripheral tissues that are involved in numerous physiological processes, and includes the two endocanabinoids:[1-4]
  - Anandaminde (AEA)
  - 2-Arachidonoylglycerol (2-AG)

- At the molecular level, abnormalities seen in patients with FXS appear to be rooted in dysregulation of the endocannabinoid pathways in the central nervous system, and include:[5-7]
  - Loss of synaptic plasticity[8]
  - Anxiety[9-10]

1. Elmes MW, et al. *J Biol Chem.* 2015;290(14):8711-8721; *2.* Castillo A, et al. *Neurobiol Dis.* 2010;37(2):434-440; 3. Mouslech Z, Valla V. *Neuro Endocrinol Lett.* 2009;30(2):153-179; 4. Pacher P, et al. *Pharmacol Rev.* 2006;58(3):389-462; 5. Zhang L, Alger BE. *J Neurosci.* 2010;30(16):5724-5729.

6. Jung KM, et al. *Nat Commun.* 2012;3:1080; 7. Maccarrone M, et al. *Neuropsychopharmacology.* 2010;35(7):1500-1509; 8. Huber KM, et al. *Proc Natl Acad Sci U S A.* 2002;99(11):7746-7750; 9. Blessing EM, et al. *Neurotherapeutics.* 2015;12(4):825-836; 10. Bergamaschi MM, et al. *Neuropsychopharmacology.* 2011;36(6):1219-1226.

CBD Neurobiology and Potential ZYN002 Mechanism of Action in FXS

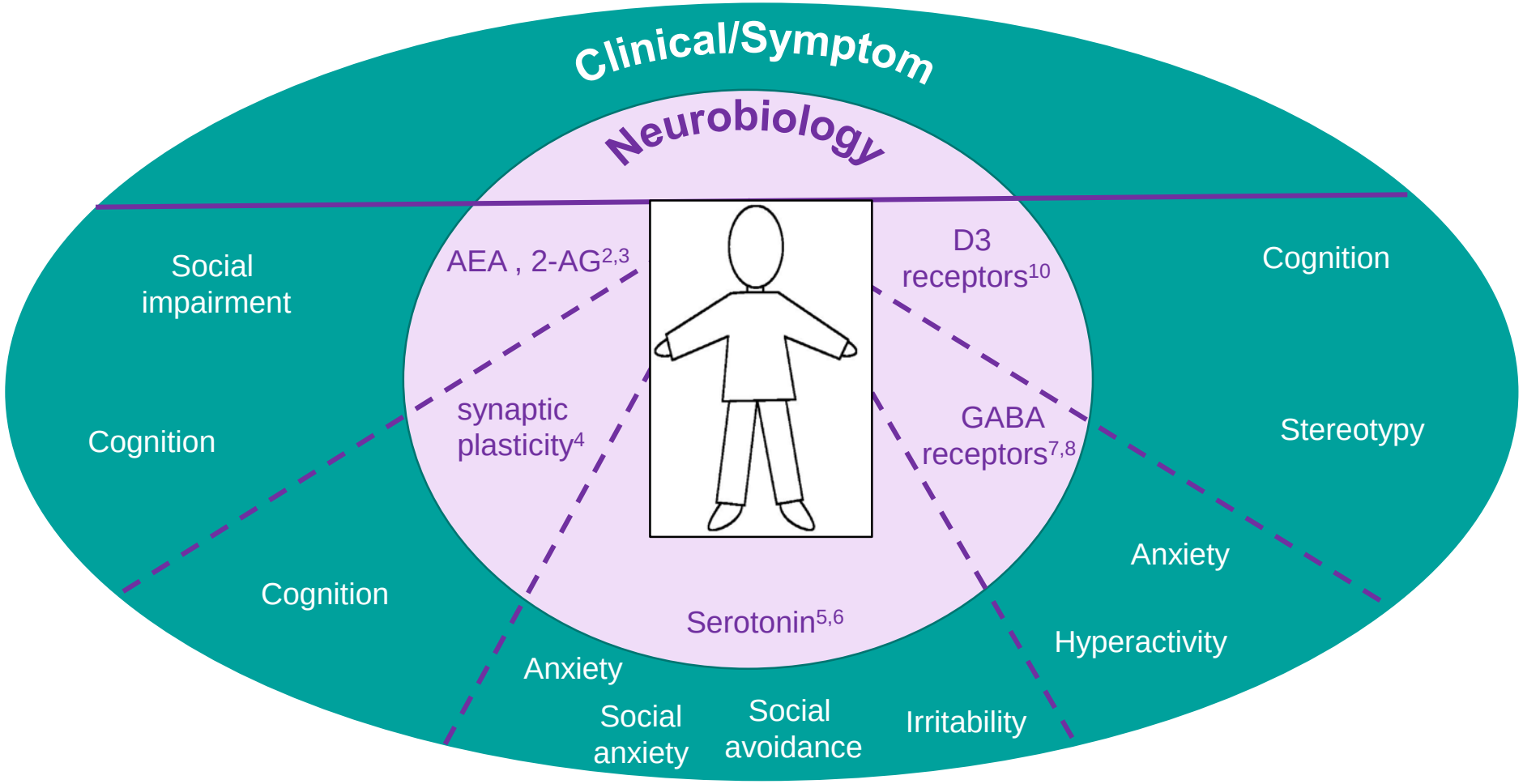
CBD effects are complex and appear to extend beyond the endocannabinoid system (ECS) - suggestive of a potential spectrum of actions [1]

- Indirectly increases AEA and 2-AG[2,3]
- Increases synaptic plasticity[4]
- Agonist at serotonin (5-HT_{1A}) receptor[5,6]
- Acts as a positive allosteric modulator of GABA-A receptors[7,8]
- Activates adenosine A_{2A}[9]
- May interact with dopamine D3 receptors[10]

1. Tartaglia N, et al. *Cannabis Cannabinoid Res.* 2019;4(1):3-9; 2. Jung KM, et al. *Nat Commun.* 2012;3:1080; 3. Maione S, et al. *Br J Pharmacol.* 2011;162(3):584-596; 4. Huber KM, et al. *Proc Natl Acad Sci U S A.* 2002;99(11):7746-7750; 5. Blessing EM,. Neurotherapeutics. 2015;12: 825–836.

6. Bergamaschi MM, et al. *Neuropsychopharmacology.* 2011;36(6):1219-1226; *7. Lozano R, Curr Pharm Des. 2015;21:4972–4979; 8. Bakas T. Pharmacol Res. 2017;119:358–370;* 9. Castillo A, et al. *Neurobiol Dis.* 2010;37(2):434-440; 10. Bian Y *Acta Pharmacol Sini.*2019;40:374-86.

FXS Neurobiology and Subsequent Symptoms Includes a Number of Different Pathophysiologic Pathways



1. Tartaglia N. Cannabis Cannabinoid Res. 2019; 4(1): 3–9; 2. Jung KM, Nat Commun. 2012;3:1080; 3. Maione S. Br J Pharmacol. 2011;162: 584–596; 4. Huber KM, Proc Natl Acad Sci U S A. 2002;99:7746–7750; 5. Blessing EM,. Neurotherapeutics. 2015;12: 825–836.

6. Bergamaschi. Neuropsychopharmacology. 2011;36:1219–1226; *7. Lozano R, Curr Pharm Des. 2015;21:4972–4979; 8. Bakas T. Pharmacol Res. 2017;119:358–370;* 9. Castillo A, et al. *Neurobiol Dis.* 2010;37(2):434-440; 10. Bian Y *Acta Pharmacol Sini*.2019;40:374-86.

ZYN002 Clinical Development Program

Positive clinical findings supported further evaluation in a Phase 3 Placebo controlled study

FAB-C Open-label Phase 2 Study[1]: Completed 2017

- Twenty children and adolescents (aged 6–17 years) with a diagnosis of FXS (confirmed through molecular documentation of FMR1 full mutation)[1]
- ZYN002 transdermal gel was administered twice daily for 12 weeks, titrated from 50 mg to a maximum daily dose of 250 mg (not placebo controlled)[1]
- Statistically significant improvement from baseline to week 12 was observed in all ABC-C_{FXS} subscales, and maintained through week 116[2]
- A total of 66 TEAEs were reported in 19 patients (95%) through week 116[2]
  - All TEAEs were mild (56/66) or moderate (10/66) in severity
  - Most TEAEs were considered unrelated to study treatment (60/66)
- Treatment-related TEAEs were reported in 6 patients[2]
- One serious adverse event was reported (constipation) and was not related to treatment[2]

TEAE=treatment-emergent adverse event.

1. Heussler et al. *Journal of Neurodevelopmental Disorders*, 2019; 2. Palumbo JM 2020 American Academy of Neurology Virtual Annual Meeting, 2020.

A Randomized, Double-Blind, Placebo-Controlled Multiple-Center, Efficacy and Safety Study of ZYN002 Administered as a Transdermal Gel to Children and Adolescents with Fragile X Syndrome

CONNECT-FX: Clinical study Of CaNNabidiol (CBD) in ChildrEn and AdolesCenTs with Fragile X

CONNECT-FX

Key Aspects of the Drug Development Process

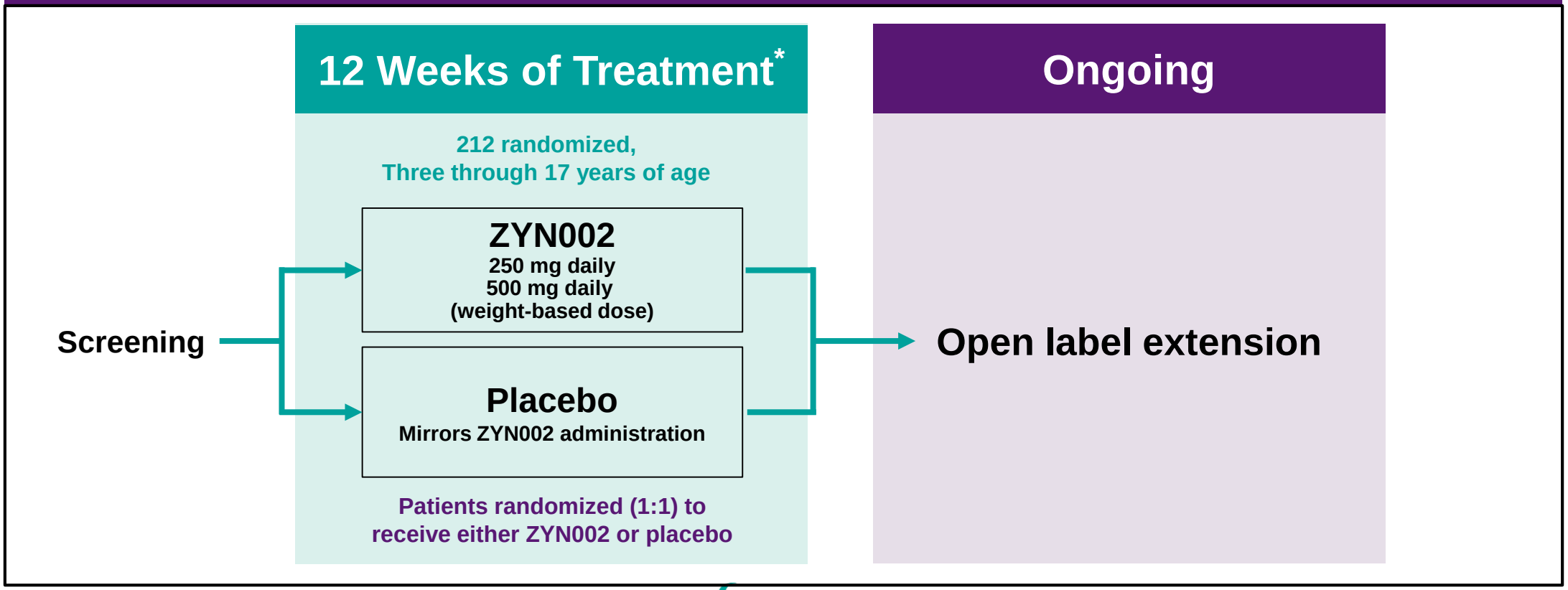
- A Phase 3 Study, often referred to as a pivotal trial, tests the efficacy and safety of an investigational drug compared to placebo
- The primary endpoint is:
  - The pre-specified finding that the study is designed to assess
  - Included in the initial statistical analysis plan filed with the FDA
- A pre-planned ad hoc analysis is:
  - Not part of the initial statistical analysis plan
  - Defined prior to unblinding the dataset

https://www.fda.gov/patients/drug-development-process/step-3-clinical-research

CONNECT-FX

Study Design

Multinational, Randomized, Double-blind, Placebo-controlled, Pivotal Study



*2 weeks placebo period, followed by 12 weeks treatment.

Sites in United States (N=17) and Australia/ New Zealand (N= 4)

CONNECT-FX

Baseline Characteristics

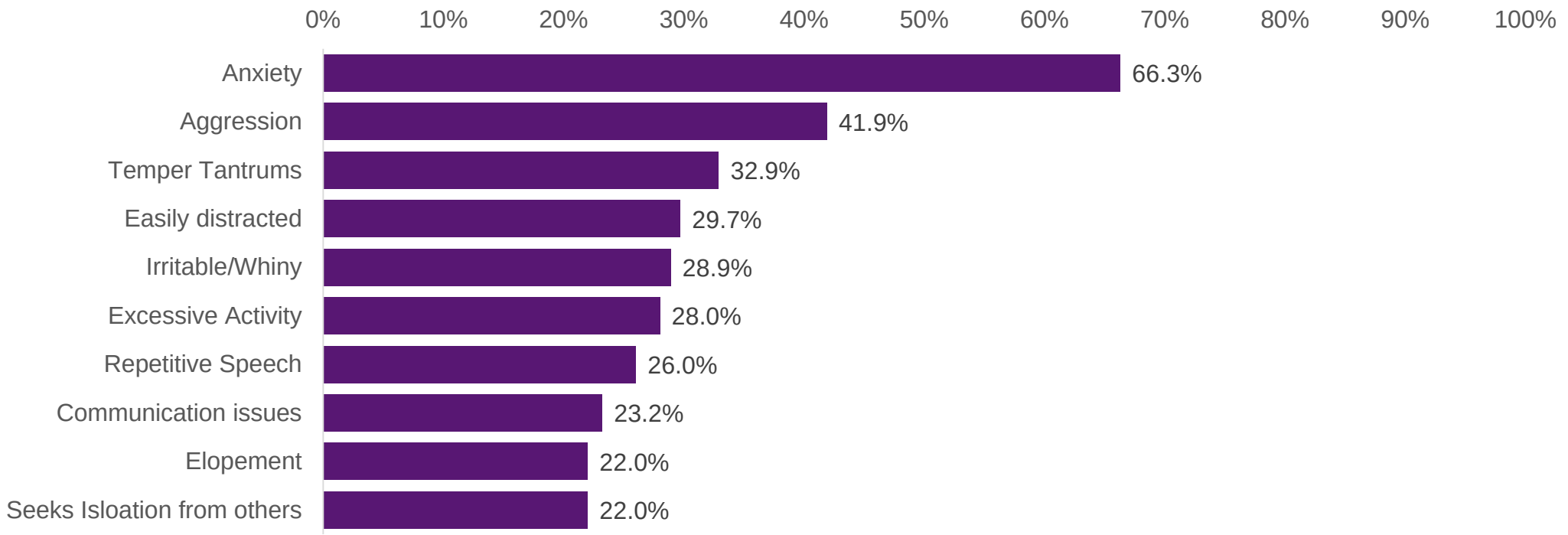
	Placebo	ZYN002	Total
n	102	110	212
Age (years)	9.8	9.6	9.7
Sex – Males, n (%)	78 (76%)	81 (74%)	159 (75%)
Weight (kg)			
Median	34.3	36.8	35.7
Range (Min, Max)	15.6, 104.7	14.6, 87.0	14.6, 104.7
>35kg, %	48.0%	55.5%	51.9%
Baseline psychoactive medications, %	66%	57%	62%

Data on File

Qualitative Caregiver Reported Behavioral Survey

Parents were asked to describe the most important behavioral challenges at baseline

Top 10 Classifications of Behavioral Challenges



CONNECT-FX

ZYN002 Did Not Meet Primary and Key Secondary Endpoints

- Primary endpoint
  - Change from baseline to end of treatment in ABC-C_{FXS} Social Avoidance subscale
- Key secondary endpoints
  - Change from baseline to end of the treatment in
    - ABC-C_{FXS} Irritability subscale score
    - ABC-C_{FXS} Socially Unresponsive/Lethargic subscale score
  - Improvement in Clinical Global Impression (CGI-I) at end of treatment, anchored to FXS behaviors

ZYN002 did not statistically significantly separate from placebo on the primary endpoint or key secondary endpoints in the full analysis set.

Pre-Planned Ad Hoc Analysis

Fully Methylated (FMet) Group (≥90% Methylation)

Not Fully Methylated (non-FMet) Group (<90% Methylation)

Rationale for Planned Ad Hoc

Building on the Scientific Evidence

Background

- DNA methylation is considered to be important in numerous pathological disorders including FXS[1]
- Methylation has been associated with the mechanism of mGluR5 in FXS[2]
- Currently, treatment options are limited for many of these disorders[1]

CONNECT-FX

- Pre-planned analysis of the most severely impacted patients defined by patients having at least 90% methylation (“full methylation”) of the impacted FMR1 gene
- Analysis to explore differences in two groups:
  - FMet group (n=167)
  - Non-FMet group (n=42)

1. Poeta L. Genes 2020, 11, 684 2014; 2. Jacquemont S. Sci Transl Med. Vol 3 Issue 64 2011.

More Severely Impacted Phenotype

Consistency with Recent Literature

Epigenetics/Methylation Status

DEVELOPMENTAL MEDICINE & CHILD NEUROLOGY — INVITED REVIEW

Epigenetics of fragile X syndrome and fragile X-related disorders

CLAUDINE M KRAAN[1,2*] | DAVID E GODLER[1,2*] | DAVID J AMOR[1,2]

1 Murdoch Children's Research Institute, Royal Children's Hospital, Parkville, Victoria; 2 Department of Paediatrics, The University of Melbourne, Melbourne, Victoria, Australia.

)–261 (1999)

FMRP Expression as a Potential Prognostic Indicator in Fragile X Syndrome

Flora Tassone,[1,2] Randi J. Hagerman,[2,3] David N. Iklé,[4] Pamela N. Dyer,[1] Megan Lampe,[2] Rob Willemsen,[5] Ben A. Oostra,[5] and Annette K. Taylor[1]

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[2]*Child Development Unit, The Children's Hospital, Denver, Colorado*
[3]*Department of Pediatrics, University of Colorado Health Sciences Center, Denver, Colorado*
[4]*Division of Biostatistics, National Jewish Medical Research Center, Denver, Colorado*
[5]*Department of Clinical Genetics, Erasmus University, Rotterdam, The Netherlands*

Phenotypic Status

RESEARCH ARTICLE

Association between IQ and *FMR1* protein (FMRP) across the spectrum of CGG repeat expansions

Kyoungmi Kim[1,2], David Hessl[1,3], Jamie L. Randol[4], Glenda M. Espinal[4], Andrea Schneider[1,5], Dragana Protic[1¤a], Elber Yuksel Aydin[1¤b], Randi J. Hagerman[1,5], Paul J. Hagerman[1,4]*

1 UC Davis MIND Institute, UC Davis Health, Sacramento, California, United States of America, 2 Department of Public Health Sciences, University of California, Davis, School of Medicine, Davis, California, United States of America, 3 Department of Psychiatry and Behavioral Sciences, University of

PEDIATRICS

OFFICIAL JOURNAL OF THE AMERICAN ACADEMY OF PEDIATRICS

SUPPLEMENT ARTICLE

Autism Spectrum Disorder in Fragile X Syndrome: Cooccurring Conditions and Current Treatment

Walter E. Kaufmann, Sharon A. Kidd, Howard F. Andrews, Dejan B. Budimirovic, Amy Esler, Barbara Haas-Givler, Tracy Stackhouse, Catharine Riley, Georgina Peacock, Stephanie L. Sherman, W. Ted Brown and Elizabeth Berry-Kravis

Pediatrics June 2017, 139 (Supplement 3) S194-S206; DOI: https://doi.org/10.1542/peds.2016-1159F

Full Data Set, FMet Group, and Non-FMet Group

Patient Disposition: 80% of Full Data Set Participants in FMet Group

Patients	Full Data Set	FMet Group	Non-FMet Group
Randomization (ITT)	212	169	42
Full Analysis set	210	167	42

- One patient did not receive study medication after randomization and one patient did not have post-baseline efficacy assessments resulting in 210 patients in Full Analysis set
- One patient with FMR1 gene deletion was not included in either the FMet or Non-FMet groups

Data on File.

CONNECT-FX: Demographics and Baseline Characteristics

Similar in the Total Analysis Group and FMet Group

	Total Analysis Group			FMet Group		
	Placebo	ZYN002	Total	Placebo	ZYN002	Total
n	102	110	212	77	92	169
Age (years)	9.8	9.6	9.7	9.6	9.2	9.4
Sex – Males	78 (76%)	81 (74%)	159 (75%)	54 (70%)	65 (71%)	119 (70%)
Weight (kg)						
Median	34.3	36.8	35.7	33.9	35.7	35.0
Range (Min, Max)	15.6, 104.7	14.6, 87.0	14.6, 104.7	15.6, 104.7	14.6, 87.0	14.6, 104.7
>35kg, %	48%	56%	52%	46%	53%	50%
Baseline psychoactive medications, %	66%	57%	62%	65%	54%	59%

Data on File.

CONNECT-FX Results: FMet Group

Pre-Planned Ad hoc Analysis Achieved Statistical Significance on Social Avoidance: Changes From Baseline to Week 12 (ABC-C_{FXS})

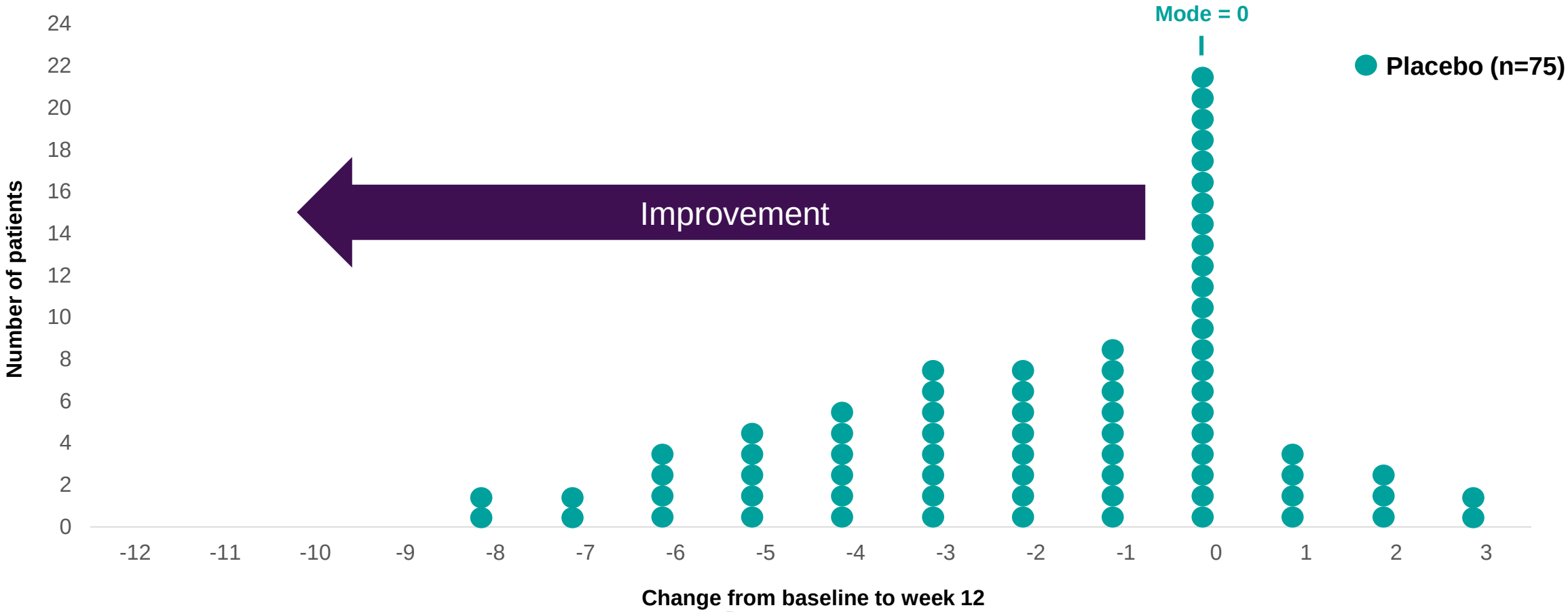
		Placebo N=76			ZYN002 N=91				
	Endpoints	Baseline Mean (SE)	Week 12 Mean (SE)	Week 12 Median Percent Change	Baseline Mean (SE)	Week 12 Mean (SE)	Week 12 Median Percent Change	Treatment Difference / Odds Ratio†	Treatment *p*-value
Primary Endpoint	**Social Avoidance**	7.18 (0.32)	5.41 (0.42)	-21.1	7.12 (0.29)	4.32 (0.33)	-40.0	-1.00	**0.020***
Secondary Endpoints	**Irritability**	28.0 (1.56)	24.11 (1.56)	-11.6	29.36 (1.37)	22.69 (1.42)	-24.3	-2.30	0.091
	Socially Unresponsive/ Lethargic	13.17 (0.85)	10.29 (0.80)	-20.5	13.30 (0.68)	9.03 (0.67)	-30.8	-1.17	0.135
	CGI-I	-	35.7%		-	51.1%		1.88†	0.056

*Statistically significant

Data on File.

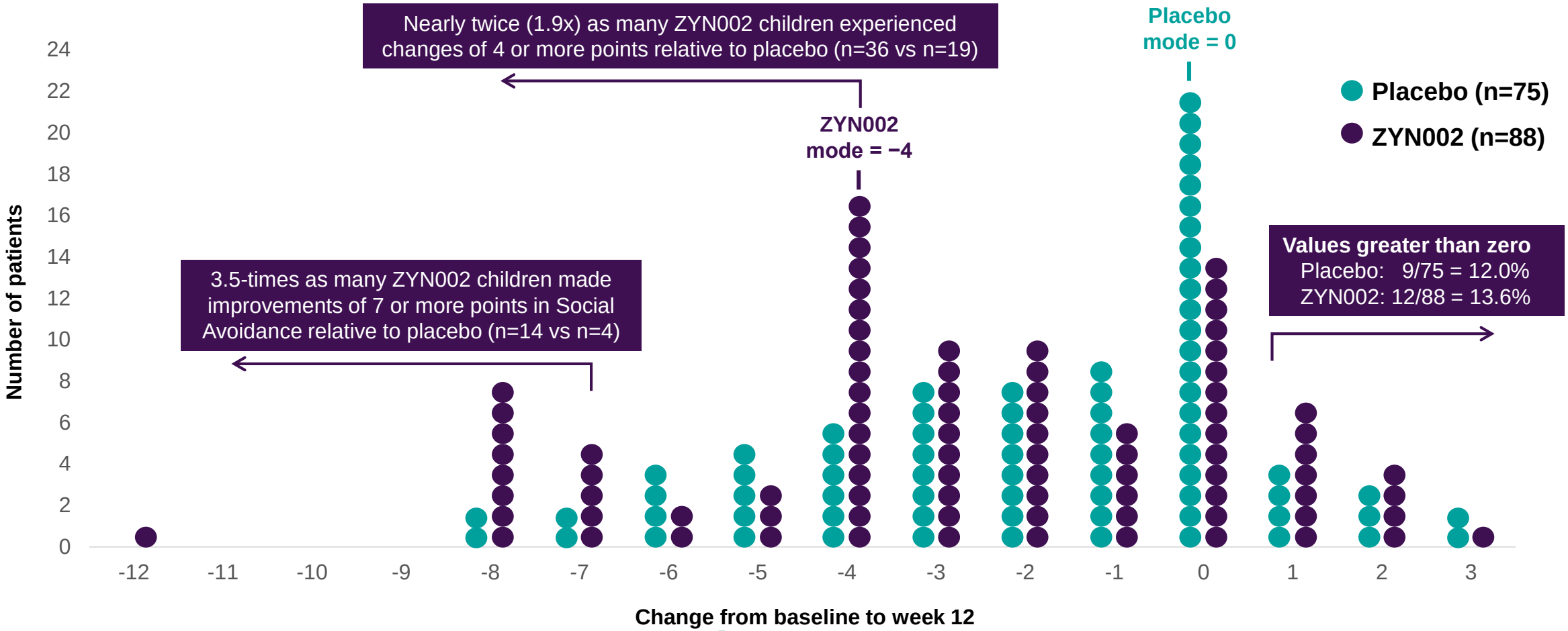
ABC-C_{FXS} Social Avoidance: Changes From Baseline to Week 12 in FMet Group

In the placebo group, the most common change (mode) in Social Avoidance was zero



ABC-C_{FXS} Social Avoidance: Changes From Baseline to Week 12 in FMet Group

The ZYN002 group, compared to placebo, demonstrated greater improvement



Data represent observed cases: 4 patients did not have Week-12 ABC-C_{FXS} assessment.

Did Caregivers See Changes in Behavior?

Caregivers were asked four questions about Impression of Change, and provided one of these seven answers:

□ Much better, □ Moderately better, □ A little better, □ No change, □ A little worse, □ Moderately worse, or □ Much worse

CAREGIVER GLOBAL IMPRESSION OF CHANGE

Question #1

- Compared to the beginning of the study, how would you rate the change in any problems your child is having with **social avoidance and isolation** (nervousness, shyness and avoidance of other people) both at home and in the community (such as at school, in stores, with family and friends)?

Question #2

- Compared to the beginning of the study, how would you rate the change in any problems your child is having with **social interactions** (communicating verbally and with “body language”) both at home and in the community (such as at school, in stores, with family and friends)?

Question #3

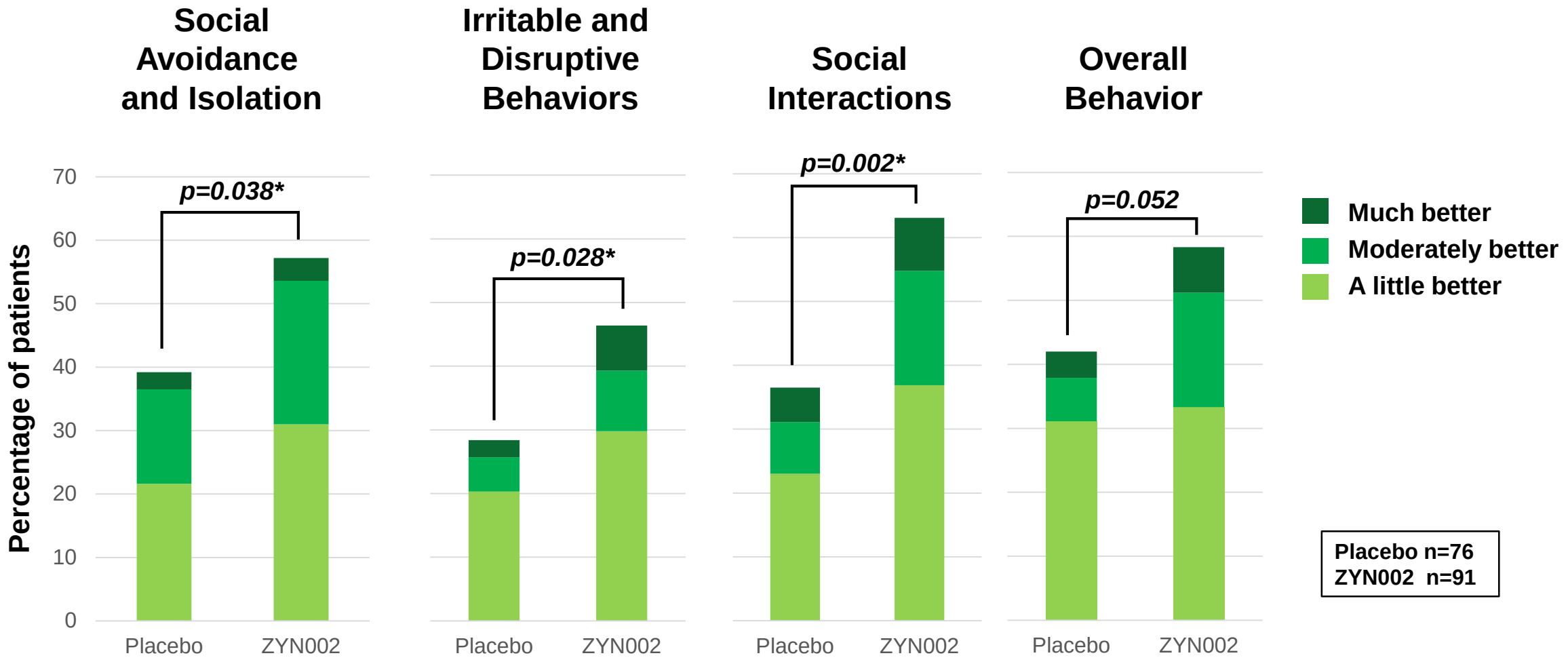
- Compared to the beginning of the study, how would you rate the change in any problems your child is having with **irritable (grumpy) behavior and disruptive** behaviors (temper tantrums, crying, whining)?

Question #4

- How would you rate the change in your **child’s behavior overall**?

Caregiver Global Impression-Change: FMet Group

Change from Baseline to Week 12: Broad Shifts Towards Global Improvement



*Statistically significant.
P-values indicate “betterment” on ZYN002 vs “betterment” on placebo.

Data on File.

Caregiver Trends Provide Observed Support for Advantage of ZYN002 in Social Avoidance (Ad Hoc FMet Group)

Clinician-rated Global Impression of Overall Improvement in Ad Hoc FMet Group Appears to be Directionally Consistent with Caregiver Observations of Social Avoidance at Week 12

ABC-C_{FXS}

- ZYN002 associated with statistically significantly improvement in Social Avoidance in FMet group
- *p*=0.020 for ZYN002 FMet compared to Placebo FMet at 12 weeks

Caregiver Global Impression - Change

- 57.2% of ZYN002 FMet patients observed to show positive change in Social Avoidance at Week 12
- 39.2% of Placebo FMet patients observed to show positive change in Social Avoidance at Week 12

p=0.038

- 63.1% of ZYN002 FMet patients observed to show positive change in Social Interactions at Week 12
- 36.5% of Placebo FMet patients observed to show positive change in Social Interactions at Week 12

p=0.002

- 46.4% of ZYN002 FMet patients observed to show positive change in Irritable and Disruptive Behaviors at Week 12
- 28.4% of Placebo FMet patients observed to show positive change in Irritable and Disruptive Behaviors at Week 12

p=0.028

Clinical Global Impression - Improvement* (anchored to FXS behaviors, and Clinician rated)

- ZYN002 FMet trended from Placebo FMet (*p*=0.056 at Week 12)

Data on File.

*Not specific to Social Avoidance

Safety Results

Safety, Tolerability, and Laboratory Assessments

CONNECT-FX

Safety

- ZYN002 was very well tolerated in CONNECT- FX, and the safety profile was consistent with previously reported clinical trials
- There were no serious or severe adverse events reported during the study
- All treatment-emergent adverse events (TEAEs) [any event, whether unrelated or related to study drug] were mild or moderate
  - The most common treatment-related TEAE was application site pain (ZYN002: 6.4%; placebo: 1.0%)
  - There were seven total psychiatric disorder TEAEs, five of which were in the placebo group
- Laboratory values for chemistry and hematology were comparable between the placebo and ZYN002 treatment groups, and there were no clinically relevant abnormalities in either group
  - There were no clinically significant changes to liver function tests

Data on File.

ZYN002 in FXS

Summary

- FXS is a heterogeneous condition and literature suggests methylation and phenotypic status influences severity and potential treatment response
- CONNECT-FX did not meet statistical significance for primary or key secondary endpoints
- In a pre-planned ad hoc analysis of FMet patients achieved statistical significance in the primary endpoint at Week 12
- Caregiver assessments for social avoidance and other behaviors showed statistically significant improvement in FMet patients
- ZYN002 was very well tolerated in CONNECT- FX, and the safety profile was consistent with previously reported clinical trials

Next Steps

These results warrant discussion with the FDA to determine path forward

Questions and Answers


Advancing Science
Zynerba®
PHARMACEUTICALS
Improving Connections
THANK YOU