

STUDY PROTOCOL WITH STATISTICAL ANALYSIS PLAN

Official title: Description of the impact of the Sentido's® Model on the adaptive behaviors of children between 3 and 7 years of age, on the autism spectrum and other associated neurodevelopmental challenges.

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SECTION I: STUDY PROTOCOL

1. SUMMARY OF THE STUDY

1.1 Background and Justification

Neurodevelopmental challenges (DNDs) affect approximately 15% of infants and adolescents worldwide, producing significant impairment in cognition, communication, adaptive behavior, and psychomotor skills. Autism spectrum (ASD) has an estimated global prevalence of 0.77%, with higher rates among males.

In Argentina, a DND rate of 20% has been reported through screening methods. Adaptive behavior, defined as the conceptual, social, and practical skills needed to function in the everyday environment, is a fundamental predictor of future quality of life and can be modified through specific evidence-based interventions.

The study aims to know the impact of the Sentido® Model on the gain of adaptive behaviors, which is important for five fundamental reasons:

1. **To improve the quality of life** of infants, families and professionals who live with neurodevelopmental challenges.
2. **Inform parents** about the efficacy and effectiveness of the interventions offered.
3. **Provide evidence** for government agencies, public and private policymakers, prepaid, and health insurance companies.
4. **Contribute** with a manualized intervention program that is contextually appropriate to the strengths and challenges of a low- and middle-income country.
5. **Provide evidence-based data** on whether the programs created fit the population needs and cultural particularities of the region.

1.2 Description of the Sentido's® Model

The Sentido's Model (SM) is an innovative proposal for the understanding and intervention of DNDs, epistemologically based on a transcomplex perspective that integrates the postulates of Morín (1996) complex thinking with Nicolescu's (1996; 2025) transdisciplinarity.

Evidence-Based Frameworks:

1. **Applied Behavior Analysis (ABA):** Interventions with extensive empirical support, recognized as evidence-based practice for ASD and DND (Hagopian *et al.*, 2015).
2. **Ayres Sensory Integration (ASI):** Validated practice for children in ASD aged 4-12 years (Schoen *et al.*, 2019).
3. **Psychoimmunoneuroendocrinology (PINE):** Stress medicine approach during critical periods of neurodevelopment (Danese & Lewis, 2017).

Operational Structure of the Sentido's® Model:

MS is a model that integrates diverse frameworks and paradigms for the intervention of children in ASD and other related DNDs, their significant caregivers and intervening therapists. To achieve this purpose, it is based on a transcomplex perspective that seeks

practical actions of a **science with consciousness** and materializes a multidimensional bio-neuro-sensory-psycho-social-spiritual-occupational-nutritional-ecological approach structured in a hierarchical architecture of four interrelated operational components.

1. NINE INTEGRATED DIMENSIONS (multidimensional framework): MS encompasses nine dimensions for comprehensive understanding, reasoning, and intervention:

Biological Dimension: Biological evolution accounts for how human behavior and genetics impact the way we act; it studies the morphophysiology and behavior of organic structures; it includes epigenetic regulation that allows the formation of different phenotypes depending on the environment.

Neurological Dimension: Examines the neurobiological aspects and functioning of the central nervous system in human development.

Sensory Dimension: The sensory processing of the central nervous system, especially the vestibular, proprioceptive, and tactile senses that impact adaptive behavior.

Psychological Dimension: The cognitive, emotional, and mental processing aspects of development.

Social Dimension: The social interactions, family dynamics, and relational contexts that influence development.

Spiritual Dimension: Integrates concepts such as ikigai (life purpose) and belief systems as therapeutic resources for psychological well-being and vital meaning.

Occupational Dimension: The systematic study of human behavior from children's play to adult work; the daily and family activities that influence health and well-being.

Nutritional Dimension: Neuronutrition as a science that links food to health and disease; it studies how nutrients affect the nervous system, behavior, and cognition.

Ecological Dimension: The study of the relationship between organisms and their environment; it includes factors such as screen time, physical activity, parental care, and social interactions that influence development.

2. SIX FUNDAMENTAL PREMISES (conceptual principles):

1. **Behavioral Premise**, which evaluates how the child and the family behave, constituting the diagnostic entry point by observing behavioral patterns and relational dynamics.

2. **Genetic/Epigenetic Imprinting premise**, which examines how genetics influences observed behavior, recognizing that family practices can modulate gene expression.

3. **Organic Condition Premise**, which identifies which medical conditions of the child (allergies, gastrointestinal disorders, sleep disturbances) impact their behavior and family functioning.

4. **Sensory Profile Premise**, which assesses how the central nervous system processes sensory information and how the family responds to these manifestations.

5. **Environmental Premise**, which analyzes how environmental, family, and social factors influence the child's development.

6. **Premise of Belief Systems and Spirituality**, which integrates spiritual strengths as therapeutic resources for the family to find meaning in the process of accompanying the child's development, generating deep and lasting systemic transformations.

3. THREE THEORETICAL POSTULATES (theoretical bases):

1. **Postulate of Neurobehavior** - Human behavior is the observable manifestation of how the central nervous system processes and integrates sensory information from the body and environment, where alterations in these processes are expressed as atypical behavioral patterns.

2. **Postulate of the Function of Behaviors** - All behavior fulfills a specific adaptive function and constitutes operant responses controlled by contingencies of three terms (antecedent-behavior-consequence) that can be identified and modified by functional analysis.

3. **Postulate of the Possibility of Conditioning Unhealthy Responses** - Maladaptive responses and problem behaviors are manifestations of alterations in the psychoneuroimmunoendocrine systems that can be modified by interventions that modulate the activation of the hypothalamic-pituitary-adrenal axis and the autonomic nervous system during critical periods of development.

4. THREE INTEGRATED STRATA (levels of intervention):

Stratum I: Childhoods in ASD and other associated DNDs - The central core that encompasses childhoods with autism spectrum characteristics and other neurodevelopmental challenges, focusing on teaching strategies to gain adaptive skills.

Stratum II: Meaningful Caregivers – Parents and caregivers, recognizing that child development occurs within family systems where caregivers' responsiveness is critical to adaptive functioning.

Stratum III: Therapists (Professionals) Involved - The health and education professionals who implement the model, requiring specialized training and interdisciplinary and transdisciplinary collaborative work.

This operational structure reflects the complexity and conceptual richness of the Sentido Model,[®] providing the complete theoretical framework that supports the entire transcomplex intervention methodology. In order to teach infants to use strategies that contribute to the gain of adaptive behaviors/skills through a guided, organized, and systematic process, evidence-based, and goal-driven by goals that families consider meaningful to achieve greater well-being in children's daily lives and family dynamics, considering the holistic integration of the nine dimensions, based on the six premises and the three theoretical postulates.

2. OBJECTIVES

2.1 Main Objective

To describe the impact of the Sentido® Model on the adaptive behaviors of children between 3 and 7 years old, on the autism spectrum and other associated neurodevelopmental challenges, who attend the Sentido's Therapeutic Organization, during a period of 17 weeks.

2.2 Secondary Objectives

1. Implement the syllabus protocol and MS programs in participating families
2. Apply protocols in children between 3-7 years of age with associated ASD and DND
3. Evaluate agreed goals using Goal Attainment Scaling (GAS) at three time points
4. Identify domains and subdomains of acquired adaptive behaviors
5. To analyze results of the Family Outcome Survey (FOS) in significant caregivers
6. Perform comprehensive statistical analysis of the results obtained

3. HYPOTHESIS

The Sentido's® Model provides tools that promote and develop adaptive behaviors in children in the AE and other associated DNDs, simultaneously enhancing the competencies, confidence and care skills of families towards their children.

4. STUDY DESIGN

4.1 Type of Study

Prospective longitudinal intervention study, pretest-posttest design with follow-up. Non-concurrent single-subject design, multiple baseline, repeated measures between subjects.

4.2 Design Justification

This design constitutes a useful first step in the research of treatment efficacy, seeking to establish a relationship between individualized intervention and change in expected results. The subject acts as his or her own control, comparing performance before, during, and after the intervention.

The non-concurrent design offers greater flexibility in clinical settings by not requiring concurrent collection of baseline data. When replicated with multiple subjects, it provides a systematic and cost-effective method for replicating results.

4.3 Duration of the Study

- **Recruitment period:** 2023-2024.
- **Duration of individual participation:** 17 weeks.
- **Active intervention:** 14 weeks.
- **Post-intervention follow-up:** 3 weeks (3 weeks without intervention from week 15 to week 17).

4.4 Study timeline

4.4.1 Schedule of general activities

Activities	2023	2024	2025
Update bibliography	✓	✓	✓
Transmit the methodology of the Sentido's Model to the participating professionals	✓	✓	✓
Select Population	✓	✓	✓
Sign consents	✓	✓	✓
Implement the syllabus protocol and the Modelo Sentido's programs in families	✓	✓	✓
Administer the scales: Vineland-3, GAS, FOS	✓	✓	✓
Apply the syllabus protocol and MS programs	✓	✓	✓
Remanage: GAS Scale, FOS Scale, Vineland-3	-	✓	✓
Statistically analyze the results obtained	-	✓	✓
Report Results Achieved	-	-	✓
Thesis writing	-	-	✓

4.4.2 Timeline of the phased study

Phase	Activity	Duration	Responsible
Preparation	Staff Training	4 weeks	PI
Recruitment	Identification and Consent	12 months	Clinical Team

Phase	Activity	Duration	Responsible
Intervention	MS Implementation	14 weeks/participant	Certified Therapists
Tracking	Post-intervention evaluations	3 weeks/participant	Evaluators
Analysis	Statistical analysis and reporting	8 weeks	Biostatistician + PI

4.4.3 Gantt Chart - Integrated Timeline

Legend of Study Phases				
●●●● PREPARATION	●●●● RECRUITMENT	●●●● INTERVENTION	●●●● FOLLOW-UP	●●●● ANALYSIS

Integrated Timeline (2023-2025)

Phase/Activity	2023Q4	2024				2025T1-T3
	Oct-Dec	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec	Jan-Sep
PREPARATION	●●●●	-	-	-	-	-
• <i>Professional training</i>	●●●●	-	-	-	-	-
RECRUITMENT	-	●●●●	●●●●	●●●●	●●●●	-
• <i>Participant identification</i>	-	●●●●	●●●●	●●●●	●●●●	-
• <i>Informed consents</i>	-	●●●●	●●●●	●●●●	●●●●	-
INTERVENTION	-	●●●●	●●●●	●●●●	●●●●	●●●●
• <i>Sentido Model App's®</i>	-	●●●●	●●●●	●●●●	●●●●	●●●●

Phase/Activity	2023Q4	2024				2025T1-T3
	Oct-Dec	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec	Jan-Sep
• <i>Regular evaluations (Vineland-3, GAS, FOS)</i>	-	••••	••••	••••	••••	••••
TRACKING	-	-	-	-	-	••••
• <i>Post-intervention evaluations</i>	-	-	-	-	-	••••
ANALYSIS	-	-	-	-	-	••••
• <i>Statistical analysis</i>	-	-	-	-	-	••••
• <i>Writing of reports and theses</i>	-	-	-	-	-	••••

Note: The schedule shows the main activities of the study. The recruitment and intervention phases may overlap since a non-concurrent single-subject design is implemented. The estimated total duration of the study is 36 months (2023-2025).

5. PARTICIPANTS

5.1 Target Population

Universe: Children aged 3-7 years with ASD and other associated DNDs who attend Sentido's Therapy Organization CAIP&CICI (Comprehensive Pediatric Care Center and Intensive Behavioral Intervention Center), Comodoro Rivadavia, Chubut, Argentina.

Sample: All eligible childhoods during the study period.

Selection: Non-probabilistic convenience, non-random, voluntary.

5.2 Eligibility Criteria

Inclusion Criteria:

1. **Informed consent signed** by the parent or guardian.
2. **Comply with the requirements** for executive admission.
3. **Meet the DSM-V** diagnostic impression criteria for neurodevelopmental disorders:

- Intellectual disability (ID); developmental global delay (MDR) or psychomotor delay (RPM).
 - Communication disorders: language disorders (SL), speech disorders, social communication disorder (SCD), childhood-onset dysfluency.
 - Autism spectrum disorder (ASD).
 - Attention deficit hyperactivity disorder (ADHD).
 - Motor developmental disorders: developmental coordination disorder (BDD), stereotypic movement disorder, tic disorders, Tourette's disorder (TT), chronic tic disorder (TT), transient tic disorder
 - Specific learning disorders (PAC).
4. **Respond to the GAS, Vineland-3 and FOS scales** in therapeutic admission.
 5. **Willingness of significant caregivers** to accompany in the intervention process, for twelve sessions according to the syllabus protocol.

Exclusion Criteria:

1. **Childhoods that exceed the age of 7 years** in the period of study.
2. **Children diagnosed with neurological or genetic diseases**, brain injury, visual, auditory or motor sensory deficits.
3. **Children who are under the guardianship of the Argentine State.**

6. INTERVENTION

6.1 Description of the Sentido's® Model

The intervention is based on a fidelity procedure based on three structural elements:

6.1.1 Syllabus Protocol Manual Operational systematization in seven procedural phases:

1. Executive admission.
2. Therapeutic admission.
3. GAS Administration.
4. Nutritional profile evaluation.
5. Therapeutic evaluation/probing.
6. Comprehensive therapeutic intervention.
7. Re-administration scales.

6.1.2 Professional Education and Training

- **Level 1:** Theoretical-technical training (7 modules).
- **Level 2:** Theoretical-experiential training (12 topics).
- **Level 3:** Interdisciplinary and Transdisciplinary Training Program (90 hours).

6.1.3 Physical Space and Materials

- Configuration according to neuroplasticity principles.
- 4 differentiated therapeutic environments.
- Materials categorized by developmental domains.

- Compliance with specific loyalty measures.

6.2 Intervention Mode

- **Duration:** 14 weeks.
- **Intensity:** 60 minutes daily, 4 days a week.
- **Modality:** Intensive comprehensive behavioral intervention.
- **Family inclusion:** Chaining and fading system in 12 structured sessions.

7. OUTCOME VARIABLES AND MEASURES

7.1 Primary Variable

Change in Vineland-3 Adaptive Behavior Composite Score (AUC) from baseline to week 17.

7.2 Secondary Variables

- Scores by Vineland-3 domains (Communication, Daily Life, Socialization).
- GAS scores for individualized goals.
- FOS scores for family competencies.
- Skill maintenance (week 15 vs. week 17).

8. MEASURING INSTRUMENTS

8.1 Vineland *Adaptive Behavior Scales, Third Edition* (Vineland-3)

Description: The Comprehensive Interview, Parent Form (IPPC) contains 502 items to measure five domains: Communication, Daily Living Skills, Socialization, Motor Skills, and Maladaptive Behavior.

Domains evaluated:

- **Communication:** Receptive (39 items), Expressive (49 items), Written (38 items) subdomains.
- **Daily Living Skills:** Personal (55 items), Domestic (30 items), Community (58 items) subdomains.
- **Socialization:** Interpersonal Relations (43 items), Play and Leisure (36 items), Coping Skills (33 items) subdomains.

Administration time: 10-40 minutes depending on the form and age.

Psychometric properties: Internal consistency coefficients ≥ 0.90 , test-retest reliability 0.64-0.92.

8.2 Goal Attainment Scaling (GAS)

Description: A method of personalized scales that allow quantifying progress towards specific collaboratively defined objectives.

Modified measuring system: 5-point scale adapted for EA and DND:

- -2 = current performance levels (baseline).
- -1 = progress.
- 0 = expected level of results.
- +1 = somewhat more than expected.
- +2 = much more than expected.

Conversion: Kiresuk-Sherman formula for standardized T-scores (M=50, SD=10).

8.3 Family Outcome Survey (FOS)

Description: Scale that measures results in competencies, confidence and skills of caring for families towards their children with special needs.

Administration: Caregiver self-report (15 minutes).

Internal consistency: $\alpha = 0.88$

9. STUDY PROCEDURES

9.1 Sampling Periods

Phase 1 - PRE-TEST (Baseline - Week 0):

- Control period during therapeutic admission.
- Vineland-3 administration, FOS to significant parents/caregivers.
- Elaboration of collaborative GAS objectives (parents-professionals).
- Complete interdisciplinary diagnostic evaluation.

Phase 2 - INTERVENTION (Weeks 1-14):

- Implementation of the Sentido's® Model according to the syllabus protocol.
- 4 weekly sessions of 60 minutes.
- Daily record of progress by professionals.
- Inclusion of caregivers (12 structured sessions), gradual fading of their participation in the sessions.

Phase 3 - POST-TEST (Week 15):

- Re-administration of Vineland-3, FOS, GAS evaluation after 14 weeks of intervention.
- Skills maintenance evaluation.

Phase 4 - FOLLOW-UP (Week 17):

- Third administration Vineland-3.
- Evaluation of GAS (parent-professional) objectives after two weeks without intervention.
- Post-intervention sustainability analysis.
- Return to usual intervention.

10. ETHICAL CONSIDERATIONS

10.1 Ethical and Legal Aspects

The study complies with:

- Approval of the Bioethics, Science and Technology Committee (CBCyT) of the University Institute in Health Sciences - H. A. Barceló Foundation.
- Principles of the Declaration of Helsinki.
- Standards of Good Clinical Practice.
- National Law 25,326 on the protection of personal data.
- ID code NCT06477666 in Clinical Trials.

10.2 Informed Consent

Information provided includes:

1. Objectives and importance of the project.
2. Study procedures in three time phases through scales administered by health professionals.
3. Evaluation scales used (Vineland-3, GAS, FOS) and their purpose.
4. **Voluntary participation** and right of withdrawal without penalty.
5. **Absolute confidentiality** - identity will not be revealed to any person.
6. **There will be no distinction** in the type of intervention offered, whether or not you decide to participate in the study.
7. Contact information: Barbara Muriel Thomas, tomasbarbara@gmail.com, 2974635050 phone.
8. Contact the Ethics Committee for inquiries and/or complaints.

Specific documents:

- **Participant Information Sheet:** Complete document with details of the study.
- **Informed Consent of Parents/Legal Representative:** Formal authorization for participation.
- **Informed Assent of the Child:** Adapted document with drawings and photos for children's understanding.

10.3 Data Confidentiality

- **Coding:** Data anonymized by coding in a research file.
- **Restricted access:** Only researchers approved with professional secrecy.
- **Secure storage:** Network computers with access by personal key.
- **Transmission to third parties:** Only encrypted data with no direct identifiable information.
- **Destruction:** All data will be destroyed/deleted once the results are published.
- **Storage:** For the time necessary for research development (estimated 24 months).
- **Rights:** Participants may exercise rights of access, rectification, limitation, deletion, portability and opposition.

SECTION II: STATISTICAL ANALYSIS PLAN (SAP)

1. STATISTICAL OBJECTIVES

1.1 Primary Objective

To evaluate the statistical change in the Vineland-3 *Adaptive Behavior Composite (ABC)* composite score from baseline (week 0) to final follow-up (week 17) in participants who received intervention with the Sentido® Model.

1.2 Secondary Objectives

1. To compare scores by domains of the Vineland-3 (Communication, Daily Life, Socialization) between the three time points.
2. Evaluate changes in GAS (Goal Attainment Scaling) scores for individualized goals.
3. To analyze changes in FOS (Family Outcome Survey) scores for family competencies.
4. Determine skill maintenance between week 15 and week 17 (non-intervention period).

2. STUDY DESIGN AND GENERAL CONSIDERATIONS

2.1 Type of Design

- **Design:** Prospective longitudinal intervention, pretest-posttest with follow-up.
- **Configuration:** Non-concurrent single subject design, multiple baseline.
- **Analysis:** Repeated intra-subject measures with three time points.

2.2 Population Analysis

Target population: n = 24 estimated participants who meet inclusion criteria and complete evaluations in the three planned time points.

Pre-established criteria for inclusion in analysis:

- Participants aged 3-7 years with a confirmed diagnosis of ASD and/or associated DND according to DSM-V
- Signed informed consent
- Completion of the 14-week intervention protocol
- Complete data in the three temporary evaluations

3. DEFINITIONS OF VARIABLES AND OUTCOME MEASURES

3.1 Primary Variable

Variable: Change in ABC Vineland-3 composite score.

Operational definition: Difference between ABC score at week 17 minus ABC score at week 0.

Measurement scale: Continuous (standard score with mean = 100, SD = 15).

Measurement time: Week 0 (baseline), Week 15 (post-intervention), Week 17 (follow-up).

3.2 Secondary Variables

3.2.1 Dominios Vineland-3

- **Communication:** Sum of subdomains Receptive, Expressive, Written.
- **Daily Life:** Sum of subdomains Personal, Domestic, Community.
- **Socialization:** Sum of subdomains Interpersonal Relationships, Play and Leisure, Coping Skills.
- **Scale:** Standard scores (mean = 100, SD = 15).

3.2.2 Goal Attainment Scaling (GAS)

- **Objectives 1, 2 and 3:** Individually defined by family and therapists.
- **Scale:** 5-point ordinal (-2 to +2).
- **Conversion:** T-scores using the Kiresuk-Sherman formula (mean = 50, SD = 10).

3.2.3 Family Outcome Survey (FOS)

- **Definition:** Family caregiving competencies, confidence, and skills.
- **Scale:** Continuous (total score).

3.3 Characterization Variables

- Age (full years at admission).
- Sex (male/female).
- Primary diagnosis (associated ASD, DND).
- Adherence to the protocol (percentage of completed sessions).

4. SPECIFIC STATISTICAL METHODS

4.1 Descriptive Analysis

4.1.1 Quantitative variables (Vineland-3, FOS):

- Arithmetic mean and standard deviation.
- Median and interquartile range (Q1-Q3).
- Minimum and maximum values.
- 95% confidence intervals for the mean.
- Graphical representation using boxplots and time profile graphs.

4.1.2 Ordinal variables (GAS):

- Absolute and relative frequencies by category.
- 95% confidence intervals for proportions.
- Median and quartile.
- Bar charts by time frame.

4.1.3 Categorical variables:

- Absolute and relative frequencies.
- 95% confidence intervals for proportions.

4.2 Inferential Analysis

4.2.1 Primary Variable Analysis

Main method: One-factor (time) repeated measures ANOVA with three levels.

- **Intrasubject factor:** Time (3 levels: week 0, 15, 17).
- **Dependent variable:** ABC Vineland-3 score.
- **Null hypothesis:** $H_0: \mu_0 = \mu_{15} = \mu_{17}$ (there are no differences between moments).
- **Alternative hypothesis:** H_1 : At least one mean differs.

Post-hoc comparisons: Bonferroni's method for multiple comparisons:

- Week 0 vs. Week 15.
- Week 0 vs. Week 17.
- Week 15 vs. Week 17.

4.2.2 Analysis of Secondary Variables

Quantitative variables (Vineland-3 Domains, FOS):

- Repeated measures ANOVA (procedure identical to primary variable).
- Bonferroni correction for multiple comparisons.
- Analysis of planned contrasts (linear and quadratic).

Ordinal variables (GAS):

- Friedman's test for repeated measures ($k = 3$ moments).
- Dunn's post-hoc comparisons with Bonferroni's correction.
- Analysis by individual objective (Objective 1, 2, and 3).

4.3 Verification of Statistical Assumptions

4.3.1 For Repeated Measures ANOVA:

Assumption of Normality:

- **Primary method:** Shapiro-Wilk test ($n \leq 50$ appropriate for $n=24$).
- **Complementary methods:**
 - Kolmogorov-Smirnov test.
 - Visual inspection using Q-Q plots.
 - Histograms with normal curve superimposed.
- **Pre-established criteria:** $p > 0.05$ to accept normality according to protocol.
- **Planned action if violated:** Logarithmic transformation or use of non-parametric methods according to protocol specifications.

Sphericity Case:

- **Method:** Mauchly test for covariance matrix sphericity.
- **Pre-established criteria:** $p > 0.05$ to accept sphericity according to protocol.
- **Planned fixes if violated:**

- Greenhouse-Geisser (epsilon < 0.75).
- Huynh-Feldt (epsilon > 0.75).
- Lower limit (very small epsilon).

Homoscedasticity Case:

- **Method:** Levene test for equality of variances.
- **Application:** A residual differences between pairs of measurements.
- **Pre-established criteria:** $p > 0.05$ to accept homoscedasticity according to protocol.

4.3.2 For non-parametric tests (GAS):

- **Independence:** Verification of the intrasubject design.
- **Ordinal scale:** Confirmation of the ordinal nature of GAS scores.

4.4 Handling Missing Data

4.4.1 Planned Primary Analysis (Completed):

- Inclusion only of participants with complete data at the three planned time points.
- Systematic description of excluded participants and reasons according to protocol.

4.4.2 Pre-established Sensitivity Analysis:

- **Planned Method 1:** Last Observed Carried Out (LOCF).
- **Scheduled Method 2:** Worst-case scenario analysis (no change assignment).
- **Protocolized comparison:** Evaluation of agreement between methods according to a pre-established plan.

4.5 Significance Criteria and Adjustments for Multiple Comparisons

4.5.1 Level of Significance:

- **Primary analysis:** $\alpha = 0.05$ (bilateral).
- **Confidence intervals:** 95%.

4.5.2 Adjustments for Multiple Comparisons:

- **Within each variable:** Bonferroni correction for post-hoc comparisons.
- **Among secondary variables:** No adjustment (exploratory analysis).
- **Number of comparisons by ANOVA:** 3 post-hoc comparisons.

Bonferroni adjustment formula: $\alpha_{\text{ajustado}} = \alpha / \text{number of comparisons} = 0.05 / 3 = 0.0167$.

4.6 Calculation of Effect Sizes

4.6.1 For Repeated Measures ANOVA:

- **Partial eta square (η^2):** To assess magnitude of effect
 - Small: $\eta^2 = 0.01$
 - Median: $\eta^2 = 0.06$
 - Large: $\eta^2 = 0.14$

4.6.2 For post-hoc comparisons:

- **Cohen's d:** For differences between pairs of means
 - Small: $d = 0.20$
 - Median: $d = 0.50$
 - Large: $d = 0.80$

Formula: $d = (\mu_1 - \mu_2) / \text{spooled}$

4.6.3 For non-parametric tests:

- **r effect:** $r = Z/\sqrt{N}$ for Friedman test
 - Small: $r = 0.10$
 - Median: $r = 0.30$
 - Large: $r = 0.50$

5. SPECIFIC ANALYSIS PROCEDURES

5.1 Planned Sequence of Analysis

Step 1: Planned descriptive analysis of baseline characteristics and systematic evaluation of missing data.

Step 2: Protocolized verification of statistical assumptions for each variable according to prespecified methods.

Step 3: Planned analysis of primary variable (ABC Vineland-3):

- Repeated measures ANOVA according to protocol specifications.
- Post-hoc comparisons conditioned to significant F.
- Preset calculation of effect sizes.

Step 4: Scheduled analysis of quantitative secondary variables (Vineland-3 domains, FOS):

- Repeated measures ANOVA for each variable according to protocol.
- Post-hoc comparisons with prespecified Bonferroni correction.

Step 5: Planned ordinal secondary variable (GAS) analysis:

- Friedman's test for each objective according to design.
- Dunn's post-hoc comparisons according to protocol.

Step 6: Preset sensitivity analysis for missing data based on specified methods.

Step 7: Planned synthesis of findings and interpretation according to pre-established criteria.

5.2 Clinical Interpretation Criteria

5.2.1 Primary Variable (AUC Vineland-3):

- **Clinically significant change:** ≥ 1.0 SD (≥ 15 points).
- **Moderate change:** 0.5-0.99 SD (7.5-14.9 points).
- **Minimum change:** 0.2-0.49 SD (3-7.4 points).
- **No change:** < 0.2 SD (< 3 points).

5.2.2 GAS Variables:

- **Complete success:** T-score ≥ 60 (level +1 or +2).
- **Partial success:** T-score 50-59 (expected level 0).
- **No change:** T-score 40-49 (level -1).
- **Setback:** T-score < 40 (level -2).

6. STATISTICAL SOFTWARE AND QUALITY CONTROL

6.1 Software

- **Main program:** SPSS version 30.0
- **Add-on software:** Microsoft Excel for data management
- **Version control:** Daily database backup

6.2 Planned Data Quality Control

- **Programmed double keying:** For 10% of randomly selected data
- **Systematic Range Checking:** Identifying Values Outside Expected Ranges
- **Protocolized Internal Consistency:** Checking for Consistency Between Related Variables
- **Outlier detection:** Identification by preset Tukey method ($IQR \times 1.5$)

6.3 Planned Analysis Documentation

- **SPSS Syntax:** Preset Command File to Use
- **Output Log:** Scheduled documentation of all statistical outputs
- **Methodological decisions:** Systematic recording of all decisions according to protocol

7. ADDITIONAL SENSITIVITY ANALYSIS

7.1 Analysis by Planned Exploratory Subgroups

- **Age:** Programmed comparison between group 3-5 years vs. 6-7 years.
- **Diagnosis:** Pre-established ASD vs. other DNDs.
- **Adherence:** Planned comparison $\geq 80\%$ vs. $< 80\%$ of completed sessions.

7.2 Scheduled Influential Case Analysis

- **Pre-established methods:** Cook distance, standardized residuals.
- **Predefined criteria:** Values > 3 SD considered atypical.
- **Planned analysis:** Repetition of main analyses excluding influential cases according to protocol.

8. HANDLING DEVIATIONS FROM THE PLAN

8.1 Minor deviations

- Adjustments to assumption verification methods.
- Changes in graphical methods of presentation.

- **Documentation:** In statistical report.

8.2 Major deviations

- Changes in primary analysis methods.
- Modifications in variable definitions.
- **Approval:** Required from the principal investigator.
- **Documentation:** Protocol amendment.

9. STATISTICAL ANALYSIS SCHEDULE

9.1 Data Cleansing Phase (Week 1-2)

- Completeness and consistency verification.
- Identification and resolution of discrepancies.
- Final database preparation.

9.2 Descriptive Analysis Phase (Week 3)

- Basal characteristics.
- Verification of statistical assumptions.

9.3 Inferential Analysis Phase (Week 4-5)

- Primary variable analysis.
- Analysis of secondary variables.
- Sensitivity analysis.

9.4 Interpretation Phase (Week 6)

- Synthesis of results.
- Preparation of tables and figures.
- Statistical report writing

10. RECOGNIZED STATISTICAL LIMITATIONS

10.1 Design Limitations

- **Absence of a control group:** Limitation for causal inferences.
- **Intrasubject design:** Possible effects of practice or time.
- **Sample size:** n=24 limits statistical power for subgroup analysis.

10.2 Instrument Limitations

- **Local validation:** Vineland-3 and FOS not validated in the Argentine population.
- **Interpretation:** Caution required in absolute magnitudes.
- **Strategy:** Focus on consistent patterns of change.

10.3 Considerations of Multiple Comparisons

- **Secondary variables:** Increase in type I error without global adjustment.
- **Interpretation:** Secondary outcomes such as exploratory/hypothesis generators.

ANNEXES

ANNEX A: CONSENT AND ASSENT DOCUMENTS

Official informed consent and assent documents include:

1. **Participant Information Sheet:** A detailed document that explains all aspects of the study.
2. **Informed Consent of Parents/Legal Representative:** Formal document of authorization.
3. **Informed Assent of the Child:** Document adapted with visual elements for child understanding.

These documents guarantee:

- Full understanding of the study by parents/caregivers.
- Voluntary participation without coercion.
- Protection of the rights of participating minors.
- Compliance with international ethical standards.
- Confidentiality and protection of personal data according to Argentine regulations.