

**A RANDOMIZED SINGLE-DOSE THREE TREATMENT PARALLEL-GROUP  
DOUBLE-BLIND PLACEBO CONTROLLED STUDY TO EVALUATE THE  
EFFICACY OF SYNTHETIC HUMAN SECRETIN FOR THE TREATMENT OF  
AUTISM AND RELATED PERVASIVE DEVELOPMENTAL DISORDER**

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## **Abstract**

Secretin, a hormone that stimulates secretions of the pancreas, has been reported to be a treatment for autism. That role is based on a published report of three children with autistic spectrum disorder who were given porcine secretin to stimulate pancreaticobiliary secretion. Within 5 weeks of treatment, gastrointestinal symptoms were lessened and marked improvements were seen in behaviors such as eye contact, alertness, and expressive language.<sup>i</sup> Sandler has recently published the results of a double-blind, placebo-controlled trial in which 28 children with pervasive developmental disorders received one dose of 0.4 mcg synthetic human secretin (sHS)/kg. When compared to 28 subjects who received placebo, no group differences were detected.<sup>ii</sup> The goal of the present study was to assess the safety and efficacy of sHS in the treatment of autism using a double-blind placebo controlled study in which participants were followed for 12 weeks after a single infusion of study drug. Thirty children with pervasive developmental disorders (24 boys, 6 girls) were randomly assigned to either high dose (0.4 mcg/kg), low dose (0.2 mcg/kg), or placebo groups. Multiple assessments of functioning were obtained prior to the study, at 3 weeks, 6 weeks, and 12 weeks after treatment. Overall group analysis revealed that children showed general improvement in functioning regardless of treatment condition. Differential analysis, however, showed that children who were more severely affected improved over time as the dosage of secretin increased. Conversely, there also was evidence that the more mildly affected children showed some deterioration in functioning when given secretin.

## **Introduction**

The pervasive developmental disorders (PDD) are a spectrum of disorders characterized by significant deviations from normal behavior and development. Included in the spectrum is childhood autism, the symptoms of which include severe impairments in communication and social interaction as well as marked restriction of activities and interests. Autism is often further complicated by hyperactivity, sleep disturbances, aggression, eating disorders, and self-injurious behavior. Gastrointestinal (GI) symptoms including chronic diarrhea, flatulence, and constipation are observed in approximately 40% of such patients, and increased intestinal permeability has been documented.<sup>iii</sup> Significant gastrointestinal pathology has been reported by Wakefield, who has termed the inflammatory bowel disease autistic enterocolitis.<sup>iv</sup> This appears to be a panenteric disease, as Horvath has described an increased incidence of reflux esophagitis,

chronic gastritis, chronic duodenitis, and other pathology.<sup>v</sup> PDD affects four times as many boys as girls, overall affecting one in 500 individuals in the United States.<sup>vi</sup>

Secretin is a gastrointestinal hormone that has been shown to be a 27 amino acid peptide with a molecular weight of 3055.5 and an empirical formula of  $C_{130}H_{220}N_{44}O_{41}$ . The primary secretory responses to secretin in normal subjects are well documented.<sup>vii</sup> A study comparing synthetic porcine and synthetic human secretin conducted by Christ et al demonstrated equivalent kinetics and pharmacological effects in human subjects on the exocrine pancreas.<sup>viii</sup>

Synthetic human secretin (sHS) has been developed as a pure, pathogen free diagnostic agent for the evaluation of the exocrine pancreas as well as for possible therapeutic indications. The present study was conducted to ascertain the effectiveness of sHS for the treatment of pervasive developmental disorders using a randomized double-blind, placebo-controlled study.

## Method

### Participants

Patients enrolled in this investigation were children (24 boys, 6 girls) between the ages of 2 and 10, inclusive, with pervasive developmental disorders. All boys and 4 of the girls were diagnosed with childhood autism. Two girls participating in the study had the diagnosis of pervasive developmental disorder-not otherwise specified (PDD-NOS). The diagnoses were established by a developmental pediatrician, child psychiatrist, or child psychologist, based on the criteria of the American Psychiatric Association's Diagnostic and Statistical Manual of Mental Disorders, 4<sup>th</sup> edition (DSM-IV)<sup>ix</sup>. In addition, diagnostic criteria included scores greater than or equal to 31 on the Childhood Autism Rating Scale (CARS)<sup>x</sup> as well as scores greater than or equal to 90 on the Gilliam Autism Rating Scale (GARS)<sup>xi</sup>. A parent of legal guardian gave written informed consent.

None of the participants had undergone previous administration of secretin. No changes in any medication or other therapeutic modality being used to treat any of the neurodevelopmental or GI symptoms of autism or pervasive developmental disorder were made within one month of enrollment.

Children were randomly assigned to one of three groups. The high dose treatment group received a 0.4 mcg/kg of sHS, the low dose treatment group received 0.2 mcg/kg and the placebo group received saline. Synthetic human secretin 16 mcg was supplied as a lyophilized sterile powder in 10 mL vials containing 1.5 mg L-cysteine hydrochloride and 20 mg of mannitol. For the 0.2 mcg/kg dosage, the contents of each vial were dissolved in 8.0 mL of sodium chloride injection, USP. For the 0.4 mcg/kg dosage, the contents of each vial were dissolved in 4.0 mL of sodium chloride injection, USP. The placebo for this study was sodium chloride injection, USP, at a volume equivalent to that utilized for sHS. An initial intravenous test dose of 0.1 mL of their assigned dosage was administered. After observation for one minute, patients received the full, remaining dose over an additional minute. Vital signs (blood pressure, pulse rate, and temperature) were obtained prior to and 30 minutes after administration of study drug.

### **Follow-up Evaluation**

Within 3 weeks of study drug administration, patients underwent a medical history and a physical examination. Stool samples for occult blood, culture, and microscopic examination for ova and parasites were obtained. Clinical laboratory evaluation consisted of serum chemistry profile (sodium, potassium, chloride, bicarbonate, glucose, BUN, creatinine, ALT, AST Alkaline phosphate, LDH, GGT, Total Bilirubin, total protein, albumin, amylase, lipase, and ammonia); hematological profile (complete blood count, hemoglobin, hematocrit, total white blood count, MCV, MCH, MCHC, differential, and platelet count); and urinalysis. Psychometric and language measures included the Vineland Adaptive Behavior Scales (VABS)<sup>xii</sup>, CARS, GARS, Leiter International Performance Scales<sup>xiii</sup> (original or revised as clinically indicated) to measure IQ, Preschool Language Scale-3 (PLS-3)<sup>xiv</sup> – of the Test of Language Development (TOLD)<sup>xv</sup> for participants whose functioning was above 6 years 11 months.

Children returned for assessment 3 weeks after the initial administration of the study drug for the following procedures: Administration of the CARS, PLS-3, and a medical assessment including review of GI symptoms and adverse events.

At 6 and 12 weeks, the same assessments were conducted, along with the administration of the VABS, the GARS, a physical exam, a recording of any adverse events, and a repeat of the baseline clinical evaluations. At 12 weeks, children also completed the Leiter.

## Results

### Group Comparisons

A comparison of the treatment and placebo groups showed that they were similar in mean age and did not differ on baseline parental reports of severity, overall Vineland score, Leiter score, or CARS score. The placebo group had a higher baseline GARS score ( $M = 106$ ) than the low dose ( $M = 97$ ) and high dose ( $M = 98$ ) ( $p < .03$ ). Although the ratio of boys to girls was not significantly different in the three groups, relatively more girls were in the placebo group (4) than in the other treatment groups (1 each). One girl with PDD-NOS was in high dose group and the other was in the low dose group.

### Analytic Plan

The majority of the analyses involved using analysis of variances with baseline data as a covariate entered before treatment effects were assessed. We did this to control for initial levels of performance before infusion. Thus, any differences found occurred above and beyond initial levels. Each time period was analyzed separately to ascertain duration of effectiveness of treatment. Additional analyses involved assessing changes over time by including the baseline (0), 3, 6, and 12 week data in a single repeated measures analysis of variance.

### Clinical Assessments

Analysis of the CARS showed that the children in the three groups did not show significantly different patterns at 3 weeks, 6 weeks, or at 12 weeks. However, in the repeated measures analysis, a significant effect was found for time,  $F(3, 81) = 15.56$ ,  $p < .001$ , with all groups showing decreasing scores over time.

Analysis of the GARS showed similar patterns in that no significant treatment groups differences were found at 6 or 12 weeks. The groups did differ in initial baseline assessments. Scores for the placebo group were higher than the other two groups. Differences in initial baseline scores



The analysis of VABS showed no significant effects due to treatment. On the Adaptive Behavior Composite, the group effect was significant at week 12,  $F(2, 27) = 4.75, p < .02$ , indicating that the placebo and high dose groups had higher scores than the low dose group, although simple effects analyses showed no significant pair wise comparisons. On the repeated measures ANOVA, the time effect was significant,  $F(2, 50) = 7.47, p < .001$ , as was the group by time interaction,  $F(4, 50) = 2.72, p < .05$ . Simple effects analyses showed that the time effect was significant only for the placebo group (showing improvement over time).

On the Vineland Daily Living subscale, the group effect was significant for week 12,  $F(2, 27) = 4.82, p < .02$ , with the placebo and high dose groups having higher mean scores than the low dose group. The repeated measures ANOVA showed a significant group effect,  $F(2, 50) = 6.28, p < .005$ , and a marginally significant group by time interaction,  $F(4, 50) = 2.35, p = .07$ . Simple effects analyses revealed that the effect for the time was significant for both the placebo and high dose groups.

On the Vineland Socialization subscale, the group effect was significant for week 12,  $F(2, 72) = 3.75, p < .05$ . On the repeated measures ANOVA, the time effect was significant,  $F(2, 50) = 13.74, p < .001$ , and the group by time interaction was marginally significant,  $F(4, 50) = 2.29, p = .07$ . Simple effects analyses showed that all three groups improved over time, although the means suggest that the placebo group showed the greatest improvement.

For the other two subscales of the Vineland, the Communication and Motor Skills subscales, similar patterns were found. Specifically, for each subscale, the repeated measures ANOVA showed that the time effect was significant (both  $ps < .05$ ), indicating improvements over time.

Analysis of the PLS overall scores showed similar patterns to the CARS and GARS in that improvement was found over time but no treatment group differences were apparent. These patterns also were apparent in the subscale analyses. For the expressive language subscale, the analyses of the 3, 6, and 12 week data showed no group differences. The repeated measure ANOVA, however, showed a significant time effect,  $F(3, 72) = 5.44, p < .001$ , with all groups showing improvements in expressive language over time.



Auditory comprehension showed a similar pattern. None of the analyses for weeks 3, 6, or 12 were significant. On the repeated measures ANOVA, the time effect was significant,  $F(3, 72) = 11.16, p < .001$ , indicating overall improvement over time.

### **Differential Responding to Secretin as a Function of Initial Level of Severity**

One purpose of our study was to examine the extent to which children differentially responded to the infusion of sHS. To accomplish this goal, we conducted a series of hierarchical regression analyses in which we used a child's initial level of a particular measure in combination with the dosage level of sHS infused (placebo, low dose, high dose) to predict that child's level on the same measure taken 3, 6 or 12 weeks later. In addition, we included a multiplicative interaction term created by multiplying the level of dose (coded as 0, 1, or 2 for placebo, low and high dose, respectively). To minimize colinearity, each main effect was centered by subtracting the group mean from each child's score for a particular measure.<sup>xvi</sup> For example, to examine the prediction of children's scores on the CARS at 6 weeks, we first entered their baseline score on the CARS and the level of sHS received. We next entered the interaction term consisting of the product of baseline CARS scores and dosage level. When a significant interaction was found, the interaction was plotted according to the procedures identified by Aiken and West (1991).

**CARS.** When we ran the regressions for the prediction of children's scores on the CARS at 3, 6 and 12 weeks, in all cases initial baseline levels of CARS significantly predicted later scores on the CARS ( $bs = 1.09, 1.29$  and  $1.15, ps < .001$ , respectively. For CARS at 6 weeks, there also was a significant interaction of baseline CARS and dosage,  $b = -.32, p < .05$ . The plot for this interaction is presented in Figure 1 and reveal that children who initially had more autistic symptoms at baseline (e.g., High Baseline levels) showed fewer autistic symptoms at 6 weeks as the level of sHS received increased (slope =  $-3.16, t(24) = -3.24, p < .05$ ). In contrast, children who had relatively low scores on the CARS before infusion increased in autistic behavior at 6 weeks as the level of sHS increased (slope =  $3.56, t(24) = 3.14, p < .01$ ; see Figure 1.) This finding was not significant in the analysis of the 12-week CARS data.

**Vineland.** A similar pattern of findings was found for children's scores on the VABS at 6 and 12 weeks. In both cases, baseline scores on the Adaptive Behavior Composite index significantly predicted subsequent Vineland scores ( $bs = 1.05$  and  $1.16$ ,  $ps < .001$  for 6 and 12 week data, respectively). Additionally, at both 6 and 12 weeks, there was a significant interaction of baseline VABS scores with dosage level in the prediction of adaptive behavior ( $bs = -.60$  and  $-.76$ ,  $ps < .05$ , respectively). The plots of these significant interactions are presented in Figures 2 and 3 for 6 and 12 week VABS data, respectively.

Children whose symptoms of autism were more severe at baseline improved at 6 and 12 weeks as dosage level of sHS increased (slopes =  $5.82$  and  $4.89$ ,  $ts(24) = 2.81$  and  $2.60$ ,  $ps < .05$ , see Figures 2 and 3, respectively). For children with less severe symptoms at baseline, the opposite pattern was found. Impairment increased at 6 and 12 weeks as the dosage level of sHS increased (slopes =  $-8.62$  and  $12.88$ ,  $ts(24) = -3.45$  and  $4.82$ ,  $ps < .01$ ). Thus for both the CARS and the VABS, children who were initially more severely impaired showed improvements over time if they received higher levels of sHS whereas children who initially showed fewer autistic symptoms fared less well over time if they received higher levels of sHS.

### **Receptive and Expressive Language**

There are no significant findings for the analyses of the PLS-3 receptive language scales at either 3, 6, or 12 weeks. For expressive language, the only significant finding was that at 3 weeks, there was a significant effect for initial level of expressive language ( $b = 1.39$ ,  $ps < .001$ , respectively). Importantly, there was a significant interaction of these two ( $b = -.59$ ,  $p < .005$ ). This interaction is plotted in Figure 4 and inspection of this figure reveals again that children who had low baseline expressive language scores improved in expressive language at 3 weeks with greater levels of sHS (slope =  $2.62$ ,  $t(20) = 2.62$ ,  $p < .05$ ). In contrast, those children who initially had the highest levels of expressive language showed poorer performance on expressive language at 3 weeks as the dosage of sHS increased (slope =  $-8.1$ ,  $t(20) = -3.48$ ,  $p < .01$ ). These

## Discussion

The results of this study have important implications for our understanding of the efficacy of sHS in the treatment of autism. At a time when unprecedented publicity surrounding the possibility that sHS may be one of the first significant medical interventions in the treatment of pervasive developmental disorders, our findings do not support that it has the kind of pervasive positive effects on autistic symptoms that have been widely reported. Although some children with autism did appear to improve when given sHS, other children given sHS either did not improve or got worse. Moreover, the effects of sHS on autistic symptoms seem to be dose related and affect some but not all symptoms. Additionally, the findings are complicated by the fact that the timing as to when the effects of sHS occur varies depending on the precise symptom.

Generally, no significant findings were found when we compared children in the various treatment groups. For those effects that did appear in these overall analyses, the main findings pertained to an effect of time such that children in all groups generally improved. When there were group differences, it was often the placebo group that improved more than the treatment groups. These findings suggest that there was a strong placebo effect.

More refined analyses showed that there was a differential response to sHS as a function of the initial level of severity. Children who were more severely impaired showed improved expressive communication scores at 3 weeks, on the CARS and VABS at 6 weeks, and on the VABS at 12 weeks and this improvement was dose related. In contrast, children who initially were mildly affected showed a parallel dose-dependent increase in autistic symptoms. One possible explanation for this differential effect is that there may be a particular subgroup of children which is sensitive to sHS. At this point, the characteristics that distinguish this group are unknown.

We did not find evidence for the children most likely to respond to sHS were those with gastrointestinal problems, those children who were higher functioning, or those with a history of regression. None of these factors predicted positive responses in this population.

One confounding factor in the analysis of our data was that fact that although the study protocol required that participants not undertake any new treatment interventions within one month of entering this trial, two of our participants who were randomly assigned to the placebo group initiated a gluten and casein free diet just prior to the one month mark. The benefits of these dietary changes are additive over time and may have affected the outcomes of these two individuals. These dietary changes may account for some of the improvements seen in the placebo group and point out the difficulty of conducting intervention studies in children who are undergoing a variety of other therapies.

Our findings provide limited support for the efficacy of intravenous sHS in children with autism. The findings suggest that children with more severe autistic symptoms are likely to benefit most from sHS. There appears to be a dose-related response to sHS such that the responses of lower functioning children are greater with higher doses of sHS. Based on the findings of our study, it is suggested that not all children with autism will benefit from its use. In fact, some may show an increase in autistic symptoms. Further studies to determine the effects, optimal dosage and timing of sHS administration are indicated.

Figure 1: Relation of Secretin and Baseline CARS to CARS at 6 Weeks

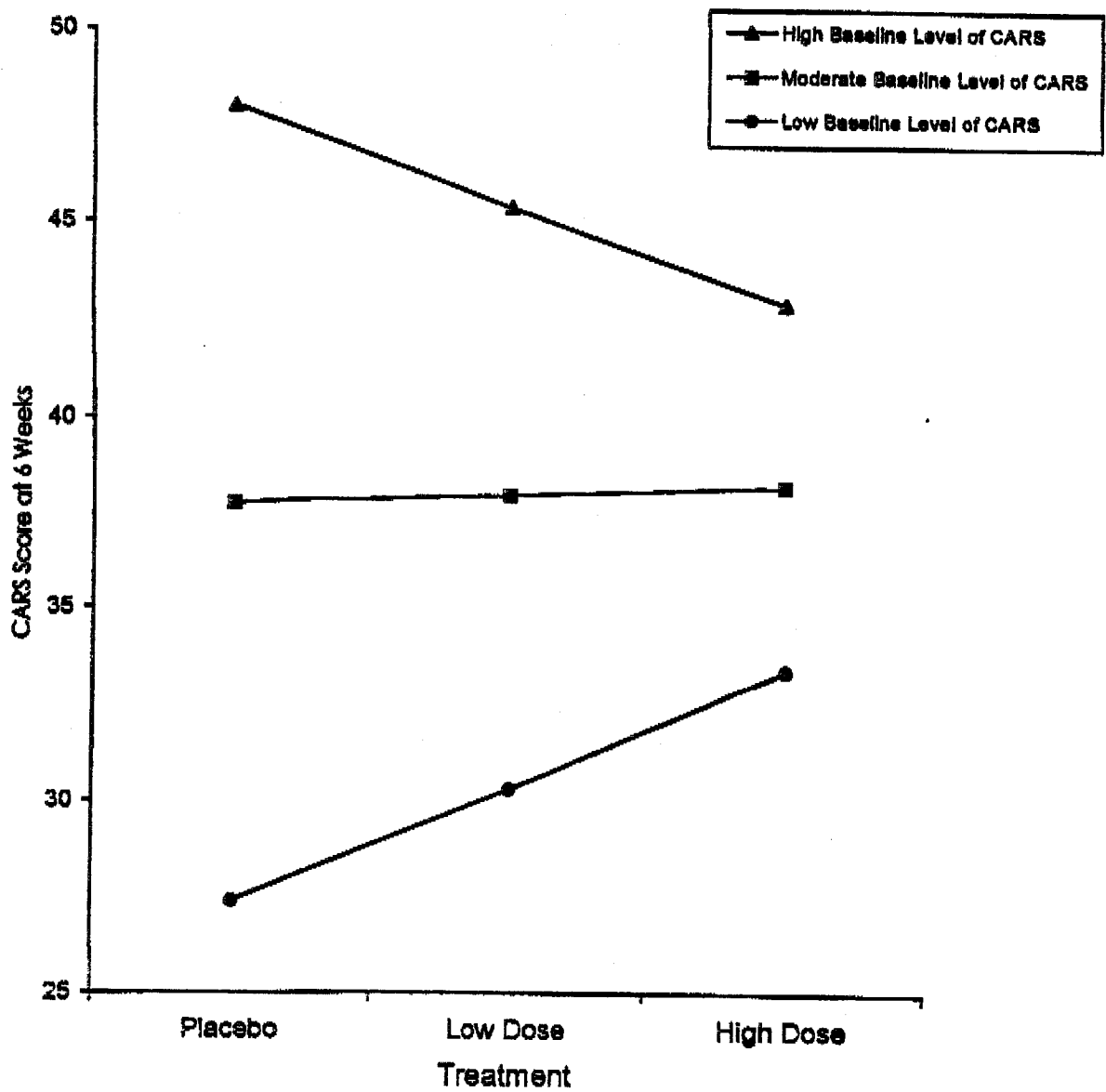


Figure 2: Relation of Secretin and Baseline Vineland Adaptive Behavior to Adaptive Behavior at 6 Weeks

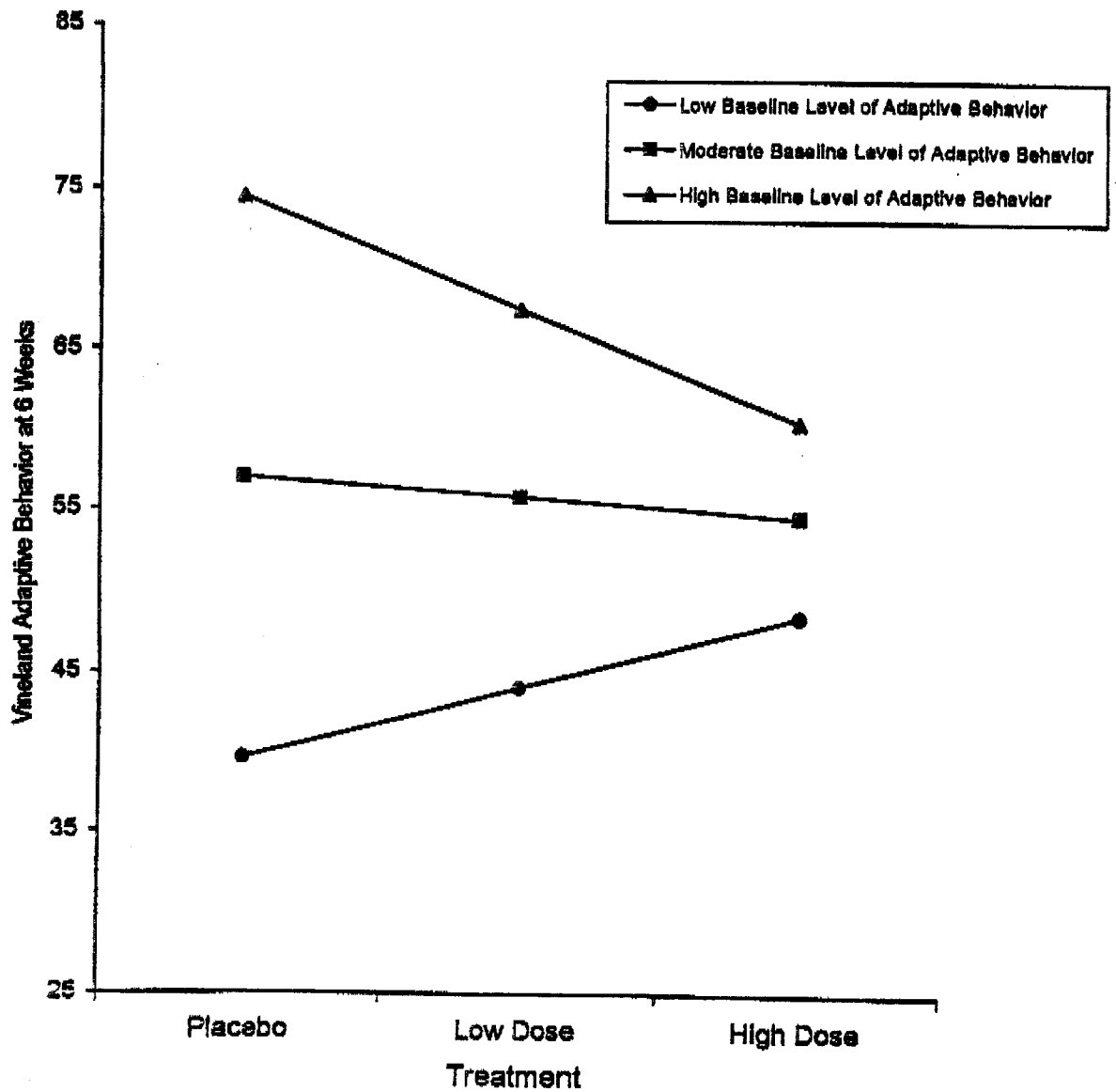


Figure 3: Relation of Secretin and Baseline Vineland Adaptive Behavior to Adaptive Behavior at 12 Weeks

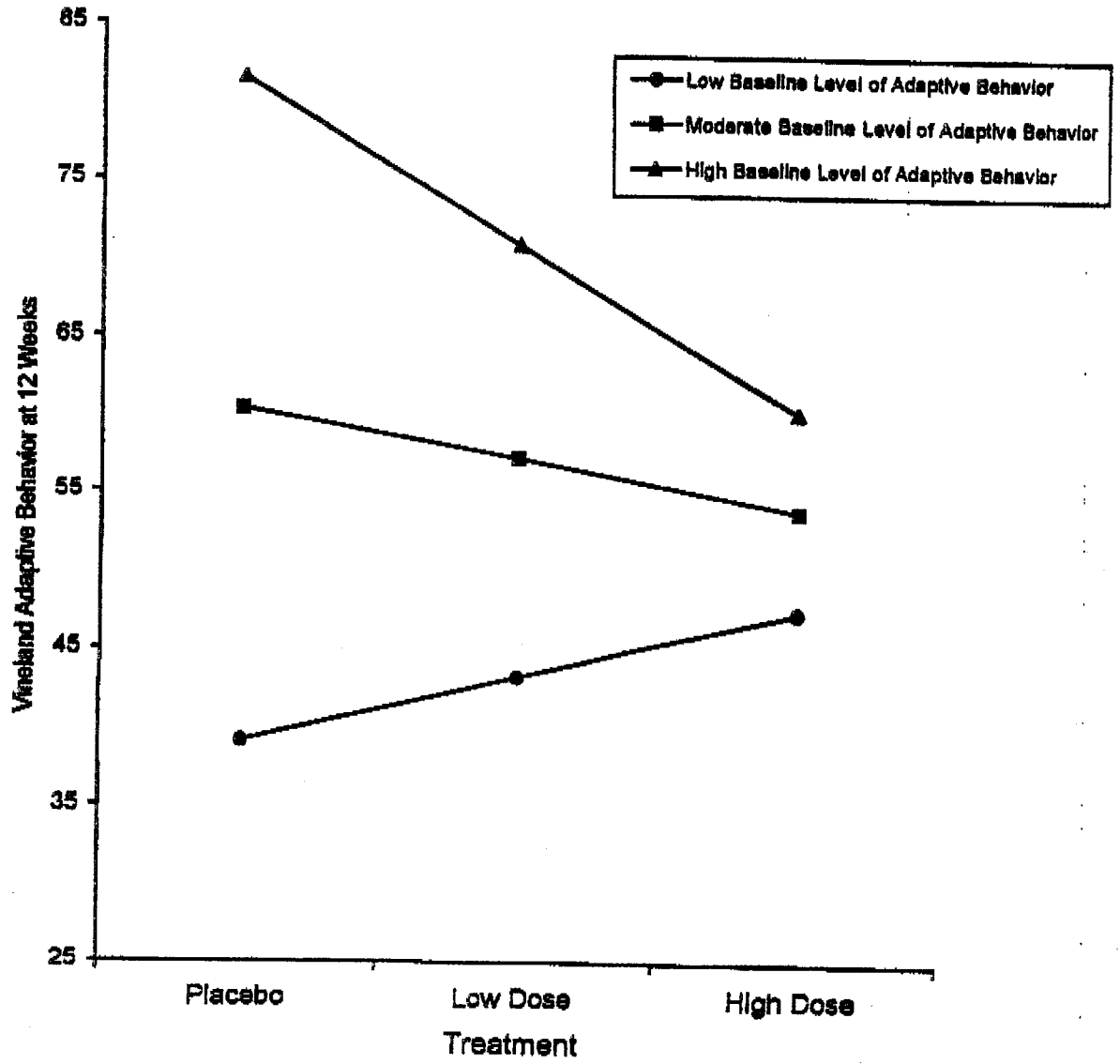
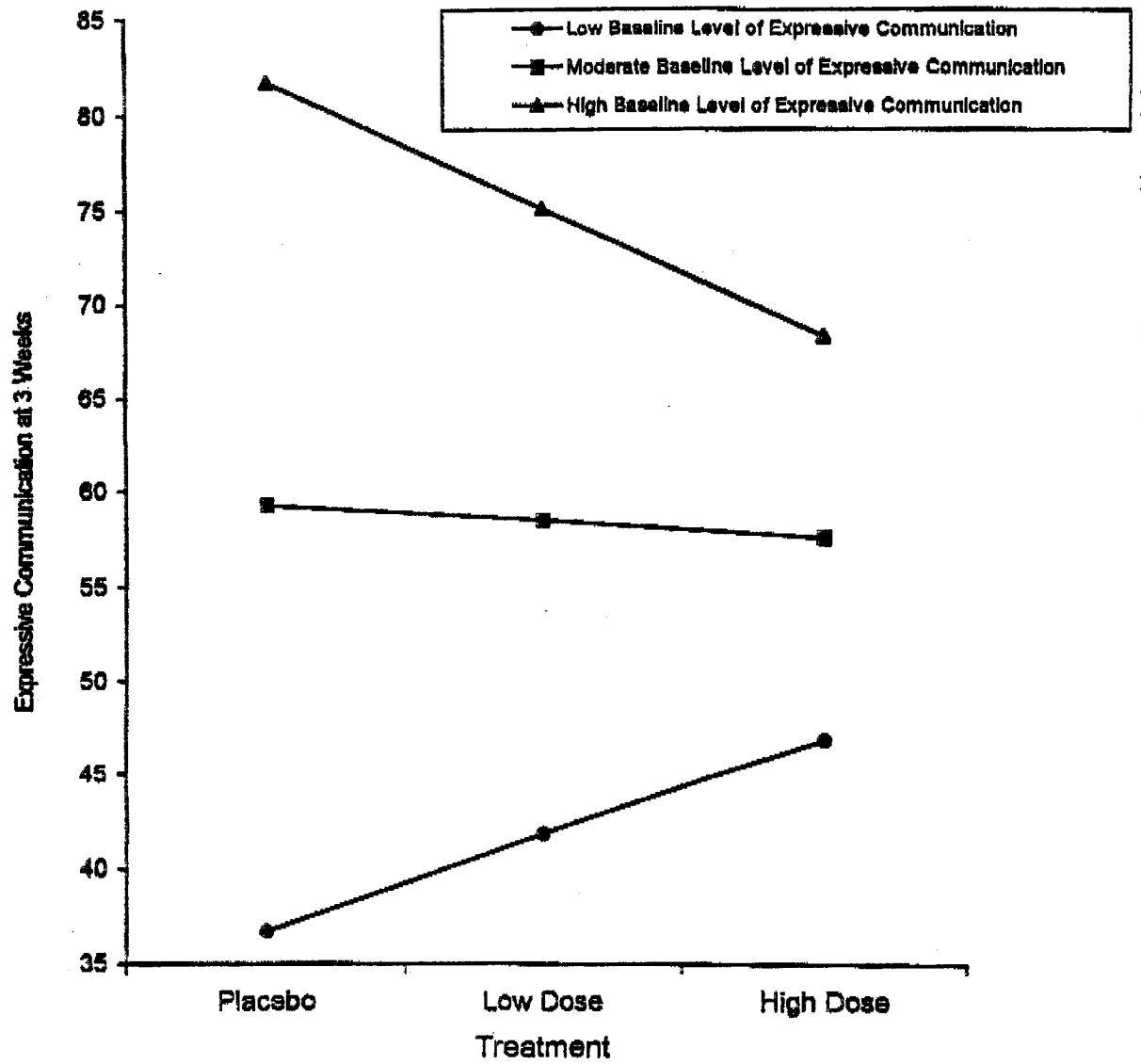


Figure 4: Relation of Secretin and Baseline Expressive Communication to Expressive Communication at 3 Weeks





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