These results are supplied for informational purposes only. *Prescribing decisions should be made based on the approved package insert in the country of prescription*			
Sponsor/company:	sanofi-aventis	**ClinialTrials.gov Identifier:**	NCT00447161
		Study Code:	ENTER_L_01125
Generic drug name:	Bacillus clausii multi-antibioresistant spores	**Date:**	17 October 2008

Title of the study:	*Efficacy and Safety of Bacillus clausii* in Preventing Antibiotic-associated Diarrhea among Filipino Infants and Children: A Multi-center, Randomized, Open-label Clinical Trial
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Study center(s):	Philippine General Hospital, Philippines De La Salle University Medical Center, Philippines

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Study period: Date first patient/subject enrolled: 17-Jul-2006 Date last patient/subject completed: 05-Oct-2007	Phase of development: Phase IV Clinical Trial
Objectives:	Primary objective: To assess the effectiveness and safety of the probiotic *Bacillus clausii* in preventing antibiotic-associated diarrhea among immunocompetent Filipino infant and children. Secondary objectives: • To assess the effectiveness of *Bacillus clausii* in reducing the incidence of diarrhea events associated with antibiotic therapy in the treatment and control group. • To assess the effectiveness of *Bacillus clausii* in reducing the gastro-intestinal (GI) related symptoms and the duration of diarrhea days among participants with AAD. • To assess the impact of *Bacillus clausii* supplementation in the over-all reduction in hospitalization days. *If a significant number is identified, we planned:* To assess the effectiveness of *Bacillus clausii* in preventing *C. difficile* associated diarrhea or colitis

Methodology:	This is a multicenter, randomised, *open-label*, clinical trial, comparing two parallel groups of infants and children allocated to either the *Bacillus clausii* probiotic strain or *a "no-intervention" control while on* antibiotic therapy on the *efficacy and safety in preventing* AAD. Restricted randomization was used in order to achieve balance between groups in size or characteristics. Blocking was used to ensure that comparison groups will be of approximately the same size. This ensured balance of the numbers in each group at any time during the trial. Complete blocks of varying sizes were randomly allocated by a "third party" through a central telephone randomization system. Each patient was identified using a center number, a treatment number (provided by the treatment code found in the intervention drug label) and the patient's initials. Investigators strived for complete separation of the people involved in the generation and implementation of assignments as well as assigning a different outcome assessor to minimize bias. Daily visits are scheduled: Day 0, then daily until day 45 post-treatment *Treatment Group* Children were treated with the B. clausii while on antibiotic therapy and shall be monitored for until the 6th week after the discontinuation of antibiotic therapy. *Control Group* Children were just observed and monitored for the development of diarrhea while on therapy until the 6th week post-discontinuation of antibiotic therapy.		
Number of patients/subjects:	Planned: 323 patients	Randomized: 323 patients	Treated: 162 patients
Evaluated: 323	Primary Outcome: • *Reduction of the incidence of antibiotic-associated diarrhea* Secondary Outcomes: • *Reduction in antibiotic-associated diarrhea events per day* • *Reduction in severity of diarrhea events* • *Reduction in GI related symptoms (nausea, vomiting, abdominal pain)* • *Reduction in hospital days* • *Reduction in C. difficile associated diarrhea or colitis* Adverse Events		
Diagnosis and criteria for inclusion:	Candidates for inclusion in the study are clinically stable infants and children: 6 months to 12 years old admitted or seen at the out-patient services in two tertiary care training hospitals (University of the Philippines-Philippine General Hospital and the De La Salle University Medical Center) for mild to moderate infection of the respiratory, genito-urinary or skin and soft tissue admitted or consulted at the outpatient for treatment of bacterial infection requiring Beta-lactam antibiotic treatment for 5 to 21 days.		

Investigational product: Dose: Administration:	1 vial of $2x10^9$ of *B. clausii* spores (BACILLUS CLAUSII) 2 billion spores of *Bacillus clausii* / 5 mL vial twice daily Per Orem	
Duration of treatment: Treatment group received a twice daily dose of 1 vial of $2x10^9$ of *B. clausii* spores *per orem* within 24 hours of antibiotic initiation and were continued until the last day of antibiotic therapy. The "No Intervention group" were only observed for events. The duration of treatment was 7 to 21 days.		Duration of observation: Participants were evaluated daily from day 0 of antibiotic therapy until day 45 post-intervention.
Reference therapy:	None	
Criteria for evaluation:		
Efficacy/Effectiveness:	Primary Outcome: • *Reduction of the incidence of antibiotic-associated diarrhea* Secondary Outcomes: • *Reduction in antibiotic-associated diarrhea events per day* • *Reduction in severity of diarrhea events* • *Reduction in GI related symptoms (nausea, vomiting, abdominal pain)* • *Reduction in hospital days* • *Reduction in C. dIfficile associated diarrhea or colitis*	
Safety:	Adverse events reported by the patient/subject or noted by the investigator.	
Statistical methods:	Data were entered in MS Access database and were analyzed using SAS V.8 software. Demographic data were described using frequencies and percentages for categorical variables. For continuous variables, mean and standard deviation were used. Differences between mean values were assessed using two-tailed Student's t test and differences between group proportions will be assessed using the x^2 test. If the sample sizes are small, differences were assessed using Fisher exact test. Efficacy will be calculated from the formula $(I_P - I_T)/I_P \times 100$, where I_P is the incidence of diarrhea in control-treated patients and I_T is the incidence in probiotic-treated patients. Relative risks were calculated from cumulative incidence ratios, and a p-value < 0.05 were defined as significant. For sub-group analysis, the plan was to assess the relative benefit of probiotic therapy in the following groups based on: antibiotic types, duration of diarrhea, hospital days and *C. difficile* associated diarrhea. To test for differences in the effect of the probiotic, a chi-square test for interaction were performed, or when appropriate a chi-square test for trend.	

Summary:

This is a multicenter, randomised, *open-label*, clinical trial, comparing two parallel groups of infants and children allocated to either the *Bacillus clausii* probiotic strain or *a "no-intervention"* control while on antibiotic therapy on the *efficacy and safety in preventing* AAD. A total of 323 infant and children were successfully randomized (Table 1.)

Table 1. Baseline characteristics of the study population

	BACILLUS CLAUSII N=162	*CONTROL N=161*
MEAN AGE (MONTHS) *RANGE*	49 (SD 34.7) 6-151	48 (SD 36.7) 6-149
SEX M:F	96: 66	94:67
MEAN WEIGHT (KG) *RANGE*	16.6 (SD 12.1) 4-132	15.9 (SD 9.7) 6-95
Mean height (cm) Range	97.3 (SD 20.0) 58-162	96.0 (SD 20.8) 65-196
Water supply NAWASA Deep well Rationed	 90 (56%) 24 (15%) 46 (29%)	 82 (52%) 27 (17%) 49 (31%)
Source of infection Respiratory Genito-urinary Skin & soft tissue	 120 (74%) 15 (9%) 26 (16%)	 119 (74%) 11 (7%) 31 (19%)
ANTIBIOTIC EXPOSURE		
Monotherapy (Number, %) Penicillins Cephalosporin Coamoxyclav/Ampicillin-Sulbactam	 69 (43) 60 (37) 12 (7)	 82 (51) 52 (32) 13 (8)
Combination/Sequential	21 (13)	14 (9)
Mean duration of antibiotic use (days, SD)	7.59 (1.86)	7.67 (2.24)

Efficacy/Effectiveness results:

Although a prevalence of 31% of children presenting antibiotic–associated diarrhea was expected in this study, only a total of 10 diarrhea events (3%) were observed (Table 2) in the included population which is extremely low to reach any conclusion.

Table 2. Episodes of diarrhea and other gastrointestinal-related symptoms

Events	Bacillus clausii n=162	No intervention n=161	Relative risk	95% CI	p-value
Diarrhea	3	7	0.43	0.11, 1.62	0.22
Nausea	1	1	0.99	0.06, 15.75	1.00
Vomiting	7	4	1.74	0.52, 5.83	0.36
Bloatedness	3	1	2.98	0.31, 28.36	0.62
Abdominal pain	3	2	1.49	0.25, 8.80	1.00

Seven (7) cases were reported in the "No intervention" group and three (3) cases in the *Bacillus clausii group* but the difference didn't reach statistical significance (p = 0.22). The frequency of other gastro-intestinal related symptoms was also low and no significant difference was observed between two groups.

Table 3. Mean age and Antibiotic Therapy of children presenting AAD

	Bacillus clausii n=3	No intervention n=7
Mean Age (SD) in months	19 (14.22) Range 9-35	21 (8.28) Range 11-32
Monotherapy (Number, %) Penicillins Cephalosporin	2 (67) 1 (33) 1 (33)	5 (71) 4 (57) 1 (14)
Combination/Sequential	1 (33)	2 (29)
Mean duration of antibiotic use (days, SD)	9 (3.21)	8 (2.24)

There was also no significant difference between the *Bacillus clausii* and "No intervention" group for secondary outcomes such as mean diarrhea episodes, duration of hospital stay and mean stool volume. There were more cases of inflammatory diarrhea (3 cases) observed in the "no intervention" arm and 1 developed *C. difficile* diarrhea. However, this did not reach statistical significance due to the unexpected low incidence of antibiotic associated diarrhea in this study (Table 4).

Table 4. Secondary outcomes among the 10 patients who developed diarrhea

DIARRHEA EPISODES	Bacillus clausii n=3	No intervention n=7	Mean difference (95% CI)	p-value
Mean number of episodes of diarrhea first 24 hrs(SD)	3.33 (0.58) Range 3-4	4.71 (1.25) Range 3-6	-1.38 (-3.16, 0.40)	0.11
Mean total number of episodes of diarrhea until resolution (SD)	13.17 (11.64) Range 5-26	15.07 (8.16) Range 4-27	-1.9 (-16.5, 12.7)	0.77
Mean duration of diarrhea (days, SD)	4.00 (3.46) Range 2-8	3.86 (2.26) Range 1-8	0.14 (-4.02, 4.30)	0.94
Stool assay results (No. of patients) Positive fecal lactoferrin Positive C. difficile toxin A/B	 1 0	 3 1		

Antibiotic-associated diarrhea is defined as otherwise unexplained diarrhea that occurs in association with the administration of antibiotics. The frequency of this complication varies among antibacterial agents. Diarrhea occurs in approximately 5 to 10 percent of patients who are treated with ampicillin, 10 to 25 percent of those who are treated with amoxicillin–clavulanate, 15 to 20 percent of those who receive cefixime, and 2 to 5 percent of those who are treated with other cephalosporins, fluoroquinolones, azithromycin, clarithromycin, erythromycin, and tetracycline (Bartlett JG N Engl J Med, 2002; Vol. 346 (5):334-339)

The prevalence of antibiotic associated diarrhea was surprisingly low in this study compared to similar population and risk factors studied elsewhere. The expected prevalence of antibiotic-associated diarrhea in this study was 31% and although, more cases were observed among children who did not receive the intervention product (Bacillus clausii) 7/10, the, low number of cases did not allow to reach statistical significance. More studies are recommended on the potential protective mechanisms inherent to the population studied on the aspect of genetic susceptibility, mucosal immune-tolerance and environmental risk and protective factors.

Safety results:	There was no trial-related adverse event observed during the course of the intervention and observation period in this study.
Date of report:	10-April-2008