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COMPARISON OF MONOSTRAIN VS MULTISTRAIN PROBIOTICS IN ACUTE WATERY DIARRHEA IN CHILDREN

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ABSTRACT

To compare mean duration of diarrhea and mean length of hospital stay between children with acute watery diarrhea treated with monostrain versus multistrain probiotics.

Place and Duration of Study: Pediatric Ward, Fauji Foundation Hospital, April to July 2026

Methodology: total of 140 children aged 6 months to 5 years admitted with acute watery diarrhea were enrolled by non-probability consecutive sampling and allocated by lottery method into two equal groups of 70. Group received monostrain probiotic (Saccharomyces boulardii) and Group B multistrain preparation, both alongside standard therapy of oral rehydration solution, zinc, and intravenous fluids where indicated. Children with persistent diarrhea, severe malnutrition, dysentery, or recent probiotic use were excluded. Baseline age, sex, weight, nutritional status, and degree of dehydration were recorded. Data were analyzed on SPSS version 26.0 using independent samples t-test and chi-square test, taking $p < 0.05$ as significant.

Results: Both groups were comparable at baseline for age, weight, nutritional status, and sex distribution ($p > 0.05$). Mean duration of diarrhea was significantly shorter in multistrain than monostrain group (2.96 ± 0.88 versus 3.57 ± 1.11 days, $p = 0.0005$), with moderate effect size (Cohen's $d = 0.61$). Mean length of hospital stay was shorter in multistrain group but not significantly so (5.12 ± 1.59 versus 5.56 ± 1.90 days, $p = 0.140$). Dehydration severity at admission was independently associated with longer diarrhea duration ($p = 0.018$).

Conclusion: Multistrain probiotic supplementation significantly shortened duration of acute watery diarrhea compared with monostrain Saccharomyces boulardii, supporting its preferential use as adjunct to standard rehydration therapy.

KEYWORDS: Acute watery diarrhea, Probiotics, Multistrain, Saccharomyces boulardii, Children, Length of hospital stay.

INTRODUCTION

Rotavirus causes most acute watery diarrhea, common and occasionally fatal infectious condition caused by bacteria, viruses, and parasites. According to WHO, diarrhea's main concern is dehydration, which happens when liquid stools, vomiting lose water and electrolytes.[1] Prolonged Recurrent Diarrhea causes malnutrition, increases risk of future health issues,[2] Subsequent infections, and nutritional deficiencies.[3] Probiotics have been widely studied as supportive treatment in pediatric diarrhea.. Efficacy is due to synergistic and complementary functional mechanisms in multiple bacterial strains, with broader pathogen cover and strong r immunomodulatory effect.[4,5]

Probiotics are increasingly recognized as useful adjuncts in management of diarrhea but current evidence base has predominantly focused on single-strain formulations, like Saccharomyces boulardii or Bacillus clausii, with scarce data directly comparing these to multistrain combinations, which could have synergistic benefits. Studies conducted in South Asia, including Bangladesh, have similarly shown significant reductions in stool frequency and diarrhea duration with multistrain probiotic use in children under five[6]. Despite widespread use of oral rehydration solutions (ORS) and advancements in public health, morbidity remains high. Probiotics, which are live microorganisms designed to enhance or restore gut microbiota, have emerged as potential intervention for acute watery diarrhea in children. Recent studies have shown that probiotics can be effective and safe, leading to reductions in duration of diarrhea and shorter hospital stays.[7, 8)]

In local study, authors reported that duration of hospitalization was shorter in children receiving multispecies probiotic group vs controls (4.94 ± 1.7 vs. 5.77 ± 1.97 days, $p = 0.002$). duration of diarrhea was significantly shorter in children receiving multispecies synbiotic group 3.25 ± 1.27 days. effect of probiotic mixture on diarrhea started after 24th hours and stool frequency significantly decreased after 24th and 48th hours. [(9)] In

another study, mean duration of diarrhea in mono-strain groups was 2.71+0.86 days, which was statistically not significant.[10]

Acute watery diarrhea remains major contributor to morbidity and mortality among children under five, particularly in low- and middle-income countries like Pakistan. question of whether multistrain probiotics lead to better clinical outcomes, such as shorter hospital stays and faster resolution of diarrhea, is of real-world importance for informing evidence-based treatment guidelines. results of this study will help optimize therapeutic choices, potentially reduce hospital burden and costs, and improve recovery rates, thereby contributing to national efforts in lowering diarrheal mortality and aligning with global child health priorities.

MATERIAL & METHOD

This was Observational Study done at Pediatric Ward, Fauji Foundation Hospital after approval of synopsis from April to July 2026. All patients of age 6 months to 5 years of either gender having acute watery diarrhea requiring hospitalization per institutional criteria; Children with no history of probiotics use in last 7 days before admission, were enrolled. And excluded were: Children with chronic or persistent diarrhea lasting more than 14 days; Children with severe malnutrition (weight-for-height Z-score < -3); Children with known immunodeficiency or chronic illnesses.; Children on concurrent antibiotic therapy other than for acute diarrhea.; having Bloody diarrhea (dysentery) or Severe electrolyte imbalance, shock, or sepsis at presentation (if present, treat and consider separate cohort). Immunocompromised (HIV, active chemo), chronic diarrheal or malabsorption disorders. Enrolled using Non probability consecutive sampling technique

Objective of study was to compare outcome of mono-strain and multi-strain probiotics in children with diarrhea in terms of mean duration of diarrhea and to compare mean hospital stay of mono-strain and multi-strain probiotics in children with severe diarrhea.

Acute Watery diarrhea: was defined as sudden onset of three or more loose or liquid stools per day, lasting for less than 14 days, and accompanied by signs of dehydration.

Dehydration status at admission was assessed using WHO clinical criteria based on physical signs and symptoms: **Mild Dehydration (Some dehydration)** was Defined by presence of two or more of following: restlessness or irritability, thirst, sunken eyes, and skin pinch that goes back slowly (less than 2 seconds); **Moderate Dehydration** was defined as Two or more of following signs: lethargy, very sunken eyes, inability to drink or drinking poorly, and skin pinch that goes back very slowly (2 seconds or more); **Severe Dehydration** was defined as Two or more of following: lethargic or unconscious state, very sunken eyes, unable to drink, or skin pinch returning very slowly (>2 seconds), often accompanied by weak or absent radial pulse and delayed capillary refill (>3 seconds); **Mono-Strain Probiotics** were Probiotic supplements containing single strain of live microorganisms, such as *Saccahomyces Boulardii*, administered in specified dose for therapeutic purposes; **Multi-Strain Probiotics** are Probiotic supplements containing two or more different strains of live microorganisms, such as combinations of *Lactobacillus* and *Bifidobacterium* species, administered in specified dose for therapeutic purposes. **Duration of diarrhea** was number of days from enrolment until passage of last watery stool, followed by at least 24 hours without diarrhea, was noted. **Length of hospital stay** was total time in days between admission and discharge.

Sample size was calculated Using WHO sample size calculation: taking expected duration of diarrhea in children receiving multispecies probiotic group (3.25 ± 1.27 days[9] vs 2.71+0.86 days[10]) receiving mono-strain probiotics; Level of significance 5%; Power of study 80%; Required sample 70 children per group (total 140).

Data for this study was collected within Pediatric Ward of Fauji Foundation Hospital after approval of institute, department and CPSP. participants were children aged 6 months to 5 years who meet inclusion criteria of having acute diarrhea.

All eligible children meeting inclusion criteria was enrolled after informed consent from parents or guardians. Baseline information including age, sex, weight, nutritional status, and duration of diarrhea before admission was recorded. Clinical assessment at enrolment will cover hydration status, stool frequency and consistency, associated symptoms such as vomiting and fever. Each child will then be randomized to receive either monostrain (group , Saccharomyces boulardii) or multistrain probiotic (group B) preparation in addition to standard therapy consisting of oral rehydration solution, zinc, and intravenous fluids as required using lottery method. study team will record total duration of diarrhea (from enrolment until last watery stool, followed by 24 hours without diarrhea) and length of hospital stay (from admission to discharge, in days). Observations was made at specific time points — at admission, after 24 hours, after 48 hours, and at discharge. At discharge, total duration of diarrhea and length of hospital stay was finalized and entered in form. Children was discharged once diarrhea has resolved, defined as no watery stools for at least 24 hours with semi-formed. Child must be clinically well, adequately hydrated, tolerating oral feeds, and have stable vital signs, as assessed by pediatrician blinded to study.

SPSS version 26.0 was used to enter and evaluate data. Baseline parameters such as age, total duration of diarrhea (days), length of hospital stay (days) was presented as means with standard deviations for continuous variables and as frequencies with percentages for categorical variables like Sex, nutritional status, degree of diarrhea. main outcome, length of diarrhea, was compared between monostrain and multistrain groups using independent samples t-test if data is normally distributed, or Mann–Whitney U test if it is not. linear regression analysis was done with age, nutritional status, and dehydration. Length of hospital stay was compared between two groups and independent sample t-test was applied. All analyses was two-tailed with significance threshold

established at p<0.05. Data of outcome variables was stratified for age, gender, severity of diarrhea, and nutritional status, and post stratification significance testing was done using independent sample t-test for continuous variables and categorical variables, taking p value of less than 0.05 as statistically significant.

RESULTS

total of 140 children aged 6 to 59 months, admitted with acute watery diarrhea, were enrolled and randomized into two equal groups: Group received monostrain probiotic (Saccharomyces boulardii, n = 70) and Group B received multistrain probiotic preparation (n = 70). All enrolled children completed study, and no participant was lost to follow-up.

two groups were comparable at baseline with respect to age, weight, nutritional status, sex distribution, and nutritional category (all p > 0.05), confirming adequate group homogeneity following randomization (Table I). distribution of dehydration severity showed trend toward more severe cases in Group B, although this difference did not reach statistical significance (p = 0.058).

mean duration of diarrhea was significantly shorter in children who received multistrain probiotic (Group B: 2.96 ± 0.88 days) compared with those who received monostrain preparation (Group : 3.57 ± 1.11 days), difference of 0.61 days (independent samples t-test, t = 3.59, p = 0.0005; Mann-Whitney U = 3251, p = 0.0008). effect size was moderate (Cohen's d = 0.61), indicating clinically as well as statistically meaningful reduction.

mean length of hospital stay was also numerically shorter in Group B (5.12 ± 1.59 days) than in Group (5.56 ± 1.90 days); however, this difference did not reach statistical significance (t = 1.49, p = 0.140; Mann-Whitney U = 2790.5, p = 0.156), with small effect size (Cohen's d = 0.25) (Table II, Figure 1). When both groups were pooled, duration of diarrhea increased progressively with dehydration severity at admission: mild dehydration 3.14 ± 1.03 days, moderate dehydration 3.18 ± 1.05 days, and severe dehydration 3.84 ± 0.90 days. One-way ANOVA confirmed this association was statistically significant (F = 4.11, p = 0.018), indicating that dehydration severity at presentation, independent of probiotic type, is important determinant of illness duration (Table III, Figure 3).

Table I. Baseline demographic and clinical characteristics of study population (N = 140)

Characteristic	Group (Monostrain) n = 70	Group B (Multistrain) n = 70	p-value
Age, months (mean ± SD)	23.0 ± 8.8	23.0 ± 10.3	0.972
Weight, kg (mean ± SD)	11.9 ± 2.0	11.9 ± 2.1	0.888
Nutritional status, WHZ score (mean ± SD)	-0.67 ± 0.96	-0.72 ± 0.91	0.797
Sex, Male, n (%)	40 (57.1)	45 (64.3)	0.489
Dehydration at admission, n (%)			0.058
Mild	30 (42.9)	37 (52.9)	
Moderate	32 (45.7)	19 (27.1)	
Severe	8 (11.4)	14 (20.0)	
Nutritional category, n (%)			0.908
Normal	45 (64.3)	44 (62.9)	
Mild malnutrition	18 (25.7)	20 (28.6)	
Moderate malnutrition	7 (10.0)	6 (8.6)	

Data presented as mean ± SD for continuous variables and n (%) for categorical variables. p-values derived from independent samples t-test (continuous variables) and chi-square test (categorical variables); p < 0.05 considered significant.

Table II. Comparison of primary outcomes between monostrain and multistrain probiotic groups

Outcome	Group (Monostrain) n = 70	Group B (Multistrain) n = 70	p-value*
Duration of diarrhea, days (mean ± SD)	3.57 ± 1.11	2.96 ± 0.88	**0.0005**
Length of hospital stay, days (mean ± SD)	5.56 ± 1.90	5.12 ± 1.59	0.140

**Independent samples t-test (both variables were normally distributed on Shapiro-Wilk testing, p > 0.05 for each group). Mann-Whitney U testing gave concordant results.*

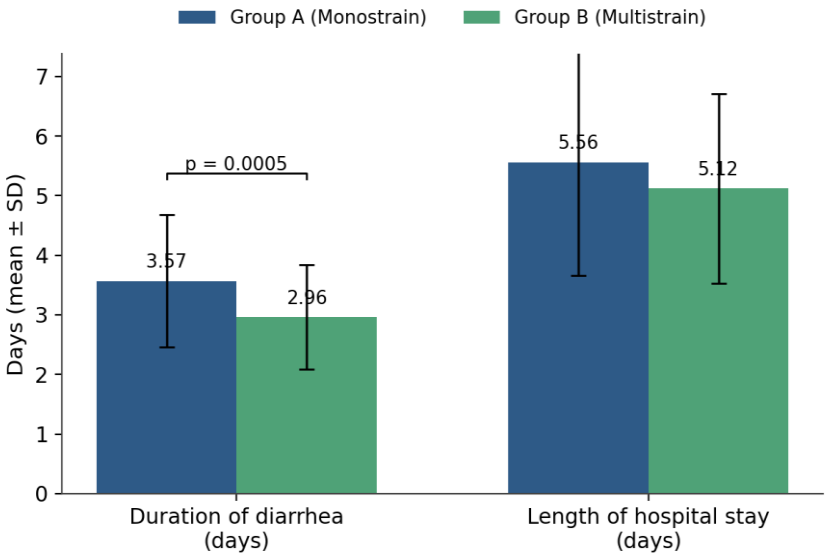

Figure 1. Mean duration of diarrhea and length of hospital stay by probiotic group (error bars represent SD).

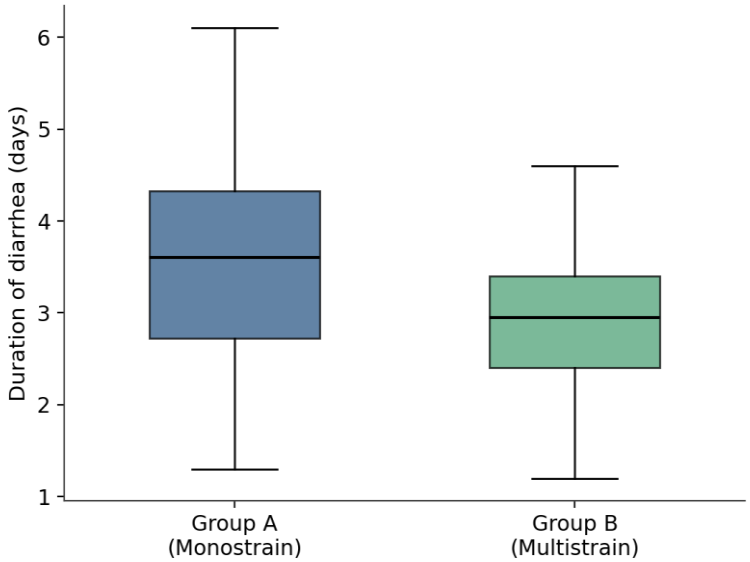

Figure 2. Distribution of diarrhea duration by probiotic group, showing median, interquartile range, and full range.

Table III. Duration of diarrhea stratified by dehydration severity (both groups combined, N = 140)

Dehydration severity	n	Duration of diarrhea, days (mean ± SD)	p-value
Mild	67	3.14 ± 1.03	
Moderate	51	3.18 ± 1.05	0.018†
Severe	22	3.84 ± 0.90	

†One-way ANOVA across three severity categories. Post-hoc pairwise comparisons were not performed given exploratory nature of this stratification.

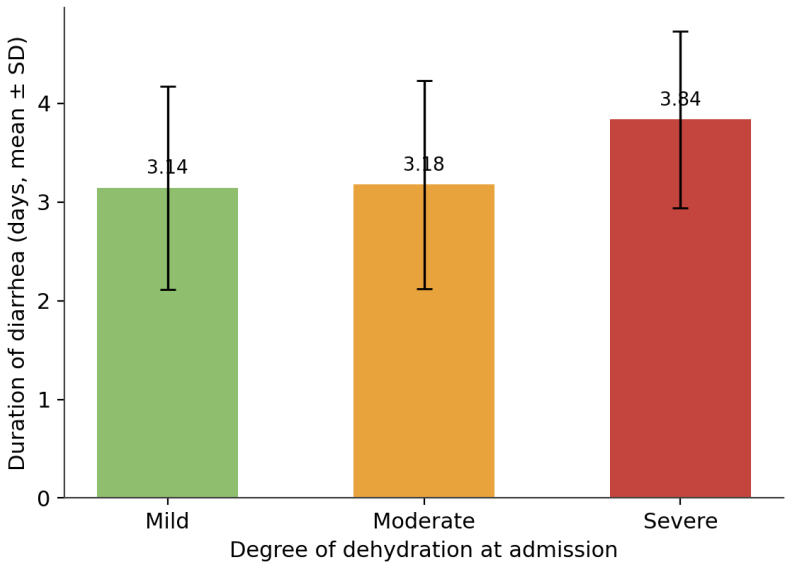

Figure 3. Mean duration of diarrhea by degree of dehydration at admission, both groups combined (error bars represent SD).

No significant linear correlation was observed between duration of diarrhea and age (r = 0.022, p = 0.795), weight (r = 0.036, p = 0.674), or nutritional z-score (r = 0.017, p = 0.843), suggesting that within this cohort, treatment allocation and presenting dehydration severity were more relevant determinants of illness duration than baseline demographic or anthropometric factors. Multistrain probiotic supplementation was associated with statistically significant reduction in mean duration of diarrhea compared with monostrain supplementation, along with shorter, though not statistically significant, length of hospital stay. Dehydration severity at presentation was independently associated with longer duration of illness.

DISCUSSION

This study compared monostrain (Saccharomyces boulardii) and multistrain probiotic supplementation in 140 children aged 6 to 59 months admitted with acute watery diarrhea. principal finding was that children receiving multistrain probiotics experienced significantly shorter mean duration of diarrhea than those receiving monostrain therapy (2.96 vs. 3.57 days, p = 0.0005), while reduction in hospital stay, though favoring multistrain group, did not reach statistical significance. These results support growing body of evidence suggesting that combining multiple probiotic strains confers greater clinical benefit than single-strain preparations in pediatric diarrheal illness.

Diarrheal disease remains leading cause of morbidity in children under five, particularly in resource-limited settings, largely through its capacity to cause dehydration and secondary malnutrition.[12,13]

Probiotics have therefore been explored extensively as adjuncts to standard rehydration therapy. Ali reported that addition of probiotics to oral rehydration solution reduced diarrhea duration compared with oral rehydration solution alone,[6] supporting general premise that probiotic supplementation, regardless of strain composition, adds measurable benefit over rehydration alone. mechanistic rationale for multistrain superiority is that different bacterial or yeast species act through complementary pathways, competitive exclusion of pathogens, mucosal barrier reinforcement, and broader modulation of mucosal immunity, so that combining strains produces additive or synergistic effects rather than relying on single mechanism of action.[4] This is consistent with meta-analytic evidence showing that probiotic mixtures outperform single-strain regimens in reducing duration of acute gastroenteritis; Huang et al. and Vassilopoulou et al. both found that multi-strain and synbiotic preparations produced greater reductions in diarrhea duration than single-strain agents across pooled pediatric trials.[7,8]

Our finding of 0.61-day reduction in diarrhea duration with multistrain therapy is broadly consistent with regional data used to power this study. Dinleyici et al. reported mean diarrhea duration of 3.25 ± 1.27 days with multispecies synbiotic mixture,[9] closely comparable to 2.96 ± 0.88 days observed in our multistrain group, while Qurrat-ul-Ann et al. reported mean duration of 2.71 ± 0.86 days with Saccharomyces boulardii monotherapy,[10] somewhat shorter than 3.57 ± 1.11 days seen in our monostrain arm. direction of effect in present study, favoring multistrain supplementation, nonetheless aligns with broader literature indicating that multistrain regimens shorten illness duration more consistently than monostrain preparations, even though absolute values differ across populations, dosing regimens, and probiotic formulations used.

absence of statistically significant difference in hospital stay between groups (5.12 vs. 5.56 days, p = 0.140) merits comment. Length of stay is influenced by numerous factors beyond diarrhea resolution alone, including feeding tolerance, associated comorbidities, parental preference, and institutional discharge practices, which may dilute treatment effect that is more clearly seen in tightly defined outcome such as diarrhea duration.

comparable pattern was reported locally, where multispecies probiotics significantly shortened hospitalization compared with controls,[9] suggesting that larger sample size or more standardized discharge protocol might be needed to detect comparable effect between monostrain and multistrain regimens specifically, rather than between probiotics and no probiotics.

Chen K, et al reported in study of 194 hospitalized children aged 1–3 years found that adding combination of three probiotic strains, Bifidobacterium lactis Bi-07, Lactobacillus rhamnosus HN001, and Lactobacillus acidophilus NCFM, to conventional rehydration therapy significantly improved acute watery diarrhoea. Compared with conventional treatment alone, probiotics reduced duration of diarrhoea by about 22.5 hours and hospital stay by 1.2 days, while also producing faster symptom improvement and less constipation.[15]

Secondary observation in our cohort was that dehydration severity at presentation was independently associated with longer duration of diarrhea, irrespective of probiotic type. This is clinically intuitive, since dehydration severity reflects both intensity of fluid losses and, indirectly, severity of underlying mucosal injury, both of which are recognized determinants of poor outcomes in diarrheal illness.[1,2] It also reinforces that probiotic supplementation should be viewed as adjunct to, rather than replacement for, timely and adequate rehydration therapy.[14,15]

Several limitations: single-center observational study ;Second, stool microbiological confirmation of causative organism was not performed, ; study duration may not have captured seasonal variation

CONCLUSION,

This study demonstrates that multistrain probiotic supplementation is associated with significantly shorter duration of diarrhea compared with monostrain Saccharomyces boulardii in hospitalized children with acute watery diarrhea, with favorable but non-significant trend toward shorter hospital stay. These findings support consideration of multistrain formulations as preferred adjunct to standard rehydration therapy, while confirming that dehydration severity at presentation remains important independent determinant of illness duration. Larger, multicenter, blinded randomized trials are recommended to confirm these findings and to clarify their effect on secondary outcomes such as length of hospital stay.

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