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Florajen®

Probiotics Reduce *Clostridium difficile* Toxin Production at Lower pH

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Introduction

The intestinal overgrowth of *Clostridium difficile*, often after disturbance of the gut microbiota by antibiotic treatment, leads to infection with manifestation ranging from mild diarrhea to life-threatening conditions. This is a serious medical problem because *Clostridium difficile* was responsible for almost half a million infections and was associated with approximately 29,000 deaths in 2011 in a study sponsored by the Centers for Disease Control. [Lessa 2015]

Methods

We evaluated the effect of pH and a probiotic called Florajen3 (15B live cultures of *Lactobacillus acidophilus, Bifidobacterium lactis* and *Bifidobacterium longum*) on toxin production by *Clostridium difficile* NAP-1 in media with 5 different pH levels before autoclaving, ranging from 5.0 to 7.0. The *Clostridium difficile* plate count and cytotoxicity titer were evaluated to identify the effect of pH on the ability of Florajen3 to inhibit toxin production.

Results

The total plate count and cytotoxicity titer for the 5 pH levels in the culture media are shown in the table below:

Medium pH before autoclaving	pH after incubation		*C. difficile* Total Plate Count	Cytotoxicity titer	
	C. diff only	*C. diff* + Florajen3		*C. diff* only	*C. diff* + Florajen3
7.0	5.5	5.5	3.4 x 10(8)	6400	400
6.5	5.5	5.3	6.6 x 10(7)	6400	<200
6.0	5.2	5.2	4.1 x 10(6)	3200	<100
5.5	5.1	5.1	1.8 x 10(6)	1600	<100
5.0	4.9	4.9	7.7 x 10(3)	<100	<100

The inoculum culture data are shown here:

Organism	pH after growth	Cell count	Cytotoxicity titer
C. difficile NAP-1	5.6	6 x 10(8)	6400
Florajen3	5.0	4.9 x 10(8)	<100

Conclusion

This study demonstrates that Florajen3 at a pH of 5.0 decreases the cell count of *Clostridium difficile* NAP-1, a particularly virulent strain, in vitro and reduces the cytotoxicity titer by almost 100-fold. Further study with Florajen3 is warranted to determine its ability to reduce the growth of *Clostridium difficile* in vivo and the associated cytotoxicity.