

Part II: Response to George Kent, World Nutrition

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Although *B. infantis* has been a part of infant nutrition for millennia, we chose to proactively assess (not assume) safety and tolerance through undertaking a well-controlled clinical trial and having an independent panel look at safety through the GRAS process.¹ Furthermore, over 40 well-controlled clinical trials have used *B. infantis* alone, or in combination with other probiotics over the past 20 years. Probiotics including *B. infantis* and other organisms have been produced routinely by well controlled fermentation, harvesting, and drying processes under principals of Good Manufacturing Practice for decades. Although some of these processes are slightly different between organisms, and may be patented, they are generally all quite similar.

The loss of *B. infantis* in the infant gut microbiome appears to be a global issue and is more limited by regional economic development and medical practice. Scientific studies from Northern Europe and Australia have indicated that although many other species of *Bifidobacterium* were detected in the gut microbiome, *B. infantis* was not observed in those studied populations.^{2,3,4} In our own work in the USA, we have published that the *Bifidobacterium* species in control babies were predominantly *B. longum* subsp *longum* and *B. breve* and in supplementary data to that publication, a table listed six *Bifidobacterium* species accounting for 100% of the total *Bifidobacterium* reads, with *B. infantis* absent from that list.⁵ This does not reflect a measurement error since 100% of the *B. infantis*-supplemented infants in this trial had a gut microbiome that was dominated by *B. infantis* to levels of over 80% of the total microbiome as long as those babies were being breastfed. Therefore, the statement that 9 out of 10 babies are missing *B. infantis* is indeed observed worldwide but limited to the highly developed nations. Finally, Henrick et al. reported a significant shift in stool pH of breastfed infants from about 5.0 in the 1920s to over 6.0 today.⁶ These are precisely the differences we were seeing in our clinical trial between infants whose gut microbiomes were colonized with *B. infantis* versus those that were missing *B. infantis*. Since the average stool pH of infant in America today is around 6.0, it is logical to conclude that most of these babies are lacking *B. infantis*.

There are likely three main reasons why *B. infantis* is missing in the infant gut microbiome today. First, when infants are fed formula rather than human breastmilk, the gut microbiome is not exposed to the full complement of human milk oligosaccharides (HMOs). Therefore, in the absence of HMOs, there is no longer a selective growth medium for *B. infantis* in the infant gut. This limits the competitive advantage naturally provided to *B. infantis* relative to other opportunistic invaders. Second, delivery by Cesarean section and modern hygienic conditions in delivery rooms significantly decreases the likelihood of the fecal-oral transfer of *B. infantis* from the mother's gut microbiome to the infant. Thirdly, and most importantly, if the mother does not have *B. infantis* herself, there is no way she can pass it on to her infant. Through generational and repeated lifetime use of antibiotics today, our gut microbiome diversity has

been significantly reduced and because of the sensitivity of *B. infantis* to antibiotics it is one of the first microbes to be lost from the mother's microbiome.^{7,8,9}

B. infantis is not a "fortifier for breastmilk" and it would be improper to think of it as such because human breastmilk already has all the nutrients required for the healthy growth of an infant up to the first 6 months of life. After that point, additional calories can be provided by combining breastmilk with complementary foods. I believe this is a general understanding that health care professionals and Evolve alike underscore. *B. infantis*, on the other hand, is naturally provided to the infant from mom's colonic flora by a fecal-oral transfer that takes place at birth. This natural transfer has likely gone on for millions of years. Ironically, without the *B. infantis* component of this symbiosis, the HMOs that make up 15% of the nutritional value of the breastmilk go unutilized by the infant and instead are excreted in the stool.¹⁰ If, however, *B. infantis* is present, this symbiosis is established, and those HMOs are converted to components (e.g., acetate and lactate) that are readily usable by the infant.⁵

Many health agencies are not necessarily at the front end of medical/scientific discovery and understanding. Indeed, the historical record is full of examples in which researcher's findings were initially dismissed, yet ultimately vindicated as the accepted doctrine (or dogma) was disproven. On May 20th, 1747, Dr. James Lind, a naval surgeon on the HMS Salisbury, undertook the first clinical nutrition experiment that ultimately established that the juice from lemons and limes significantly improved the condition known as scurvy. He had the outrageous hypothesis that the condition was a consequence of something missing in the diet (now known as Vitamin C) which was wholly disregarded by the medical establishment at the time. Rather, the common dogma at the time was that scurvy was a "*disease of internal putrefaction brought on by faulty digestion caused by the hardships of life at sea and the naval diet*".¹¹ In 1981, Dr. Barry Marshall, a young doctor in his third year of medical training in Australia noticed that S-shaped bacteria were present in many biopsy specimens from patients with ulcers and stomach cancer. In the years that followed he proposed that ulcers, long believed by the medical establishment to be caused by stress, was actually caused by the presence of *Helicobacter pylori* and that it might be cured by simple antibiotic treatment. Disregarded by medical institutions for years, he finally did a test on himself, proved his theory to be true, and ultimately shared the Nobel Prize for Medicine in 2005 with his colleague Dr. J. Robin Warren. These are just two of many examples in history where novel and simple ideas, which challenged the general medical understanding at the time, but which were based on solid scientific observations and theory, went on to magnificently impact public health. I am sure that many people, educated or otherwise, in the late 1700's and the late 1900's asked the same unanswerable question posed by Dr. Kent.

Finally, we would like to point out that although the Bill & Melinda Gates Foundation and Horizons Ventures led the last financing round for the Company, the majority of the investment came from other investors and we believe that the collective rationale for their investment is well elucidated in the publicly disclosed press release.¹²

Editor's Note: David Kyle and Tracy Shafizadeh state that they are both employees of Evolve BioSystems, maker of Evivo.

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