RESEARCH ARTICLE

Expired probiotics: what is really in your cabinet?

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One sentence summary: This study examined the viability of expired probiotic products to find that several contained live microorganisms long after the labelled expiration date.

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ABSTRACT

The popularity of using probiotics has surged, since they became widely accepted as safe and help improve general health. Inevitably, some of these products are used after expiration when microbial cell viability is below the recommended effective dose. Given that probiotics must be live microorganisms administered in adequate amounts, the aim of this study was to measure viability in expired products and assess how packaging and storage conditions impact efficacy, if at all. Thirty-three expired probiotic products were evaluated, of which 26 were stored in conditions recommended by the manufacturer. The viable microbial cells were enumerated and representative isolates identified by 16S and internally transcribed spacer rRNA gene sequencing. While the products had a mean past expiration time of 11.32 (1–22) years, 22 still had viable contents, and 5 were within or above the original product cell count claim. Product formulation and the number of species present did not appear to impact the stability of the products. However, overall packaging type, storage conditions and time since expiry were found to affect viability. All products with viable cells had the strain stipulated on the label. Despite some selected probiotic products retaining viability past their expiry date (indicating long-term storage is possible), the total counts were mostly well below that required for efficacious use as recommended by the manufacturer. Consuming expired probiotics may not yield the benefits for which they were designed.

Keywords: expired; probiotic; microbe; health benefit; packaging; storage

BACKGROUND

Probiotics are ‘live microorganisms that, when administered in adequate amounts, confer a health benefit on the host’ (Hill, Guarner and Reid 2014) and are packaged and sold for use. They can potentially offer various benefits, such as preventing antibiotic-associated and traveler’s diarrhea (Gao et al. 2010), reducing the symptoms of irritable bowel syndrome (Giannetti et al. 2017), lowering blood cholesterol (Simons, Amansec and Conway 2006) and impacting a range of other conditions ( Tanner et al. 2018). With increasing recognition of the benefits of probiotics, the global market is expected to surpass US $52 billion by 2020 (Probiotics Market Size & Share 2019). A considerable amount of product has entered the market over the
past 10 years, with presumably some accumulating in household. A survey indicated that approximately one-fifth of Americans had not tidied their personal medications within the preceding 3 years (Gill 2018), and 28% of participants in a study have unused/unwanted drugs in their homes (Akici, Aydin and Kiroglu 2018); there is potential for consumers to accumulate and use expired probiotic products.

With the accelerating costs of medications, the use of expired healthcare products has become a topical issue. Institutions that hold large stocks of prescription medicines, such as the US military, have looked at the stability of active ingredients in long-expired prescription medications (Khan et al. 2014). Over the past 35 years, hundreds of drugs have been tested and 90% of them were found to be safe and effective after expiration (Cohen 2000; Cantrell et al. 2012; Khan et al. 2014). The expiration dates of medications are often for commercial use rather than being indicative of medical potency (Cohen 2000; Cantrell et al. 2012). Vitamin supplements, for example, lack expiration studies, so manufacturer labels underestimate the amounts of active ingredients to ensure that the products contain the advertised minimum quantities at expiry (Andrews et al. 2017). In the case of probiotics, manufacturers often ‘front load’ their products to ensure a certain number of viable cells remain at the expiry date and ideally like them to be above this level for 2 years at room temperature.

As probiotics comprise live organisms, it is important to monitor handling and storing probiotics so that they do not become contaminated or degrade in a manner that makes their components harmful. While the proportion of the population willing to use probiotics after expiration has not been determined, an estimated 9.34–19.25% of people do not check the expiration date on their medicines before using them and it is probable that some of those who notice their probiotics have expired will still use them regardless (Raja 2013; Mongkolchaisapat et al. 2014). Despite this, a study has found that 51.2% of older patients comply with storage quality and information on drug labels (Vlieiland et al. 2018). A study of university students revealed they presumed that the life of medications can be extended past expiration with refrigeration (Aisoonphisarnkul et al. 2014). Furthermore, as some probiotic products are expensive (Annunziata and Vecchio 2013), consumers may be reluctant to discard them.

Probiotic products are available in a variety of forms and packages that include: refrigerated food, metal blister packs, plastic–metal sachets, capsules, and tablets in plastic and glass bottles. These formats provide varying degrees of protection from the main causes of microbial cell death under storage, namely high-water activity, oxygen and heat, yet few published scientific studies have compared packaging modalities (Miller et al. 2002; Anukam and Reid 2007; Sarkar 2010; Terpou et al. 2019). While there may be a potential benefit from the consumption of dead microorganisms, these are not classified as probiotic. Studies have proven consumption of dead, heat-treated or killed probiotics, metabolites, cell fractions, culture supernatant and probiotic microbial DNA can confer health benefit (Kataria et al. 2009; Adams 2010; Taverniti and Guglielmetti 2011). This is not surprising as anti-inflammatory and immunomodulation effects can be induced by cell-free supernatants (De Marco et al. 2018; Geraldo et al. 2020), peptidoglycans (Geraldo et al. 2020), LPS (Pique et al. 2013), exopolysaccharides (Liu 2017) and teichoic and lipoteichoic acids (Matsuguchi et al. 2003; Kim et al. 2017). This demonstrates that there may be further use for probiotic products beyond expiry (Taverniti and Guglielmetti 2011).

The goal of the present study was to assess the viability of randomly acquired, expired probiotic products and look for correlations with packaging, storage conditions and the time since expiry. We hypothesized that commercial probiotics stored in compliance with the manufacturer’s guidelines will contain viable microorganisms past the date of expiration. Where possible, the microorganisms present in each probiotic product were identified by DNA sequencing to ensure that the manufacturers had labeled the products correctly.

MATERIALS AND METHODS

Test products

Thirty-three products listed in Table 1 were tested, of which 23 are shown in Fig. 1A. They had been acquired over a number of years and stored in sealed containers at room temperature or under refrigeration.

Study outline

The following information was collected for each probiotic product evaluated: genus and species of bacteria, minimum colony forming units (CFU), packaging type, storage recommendations, expiration date and storage conditions. Products without listed expiration dates were excluded from the study.

One dose (capsule, lozenge, powder, etc.) of the probiotic product was added to 10 mL of sterile phosphate-buffered saline (PBS pH 7.4, 0.01 M). If the dosage format was encapsulated, the powder was added to the PBS; if there was no capsule, the whole contents were added. The mixture was vortexed until homogeneous and serially diluted in PBS; 5 μL of the first through sixth dilutions was inoculated, drop plate-wise, on various types of selective media as described as follows. When sufficient quantities of the probiotic product were available, six capsules were tested, each with four replicates. Otherwise, four replicates of all available doses were prepared in this manner. All plates were incubated both aerobically and anaerobically in jars at 37 °C for up to 48 h. All plates were checked at 24 h and counted if there was growth; however, if no growth was observed, plates were returned to grow for an additional 24 h. The remaining bacterial content was then calculated based on recoverable CFU compared with guaranteed CFU. Microbial contents of the probiotics were confirmed when possible with selective media based on the manufacturer’s claimed microbial contents. When this was not possible, 16S rRNA gene sequencing, and internally transcribed spacer (ITS) region sequencing for yeast were used to confirm microbial contents.

Selective media for enumeration

Several selective media were used to isolate all possible species from each probiotic. Streptococcus salivarius was grown on Mitis Salivarius Agar (Difco, Sparks, MD, USA) aerobically at 37 °C. SF Broth (20.0 g of tryptone, 5.0 g of dextrose, 4.0 g of dipotassium phosphate, 1.5 g of monopotassium phosphate, 5.0 g of sodium chloride, 0.5 g of sodium azide, 32.0 mg of bromocresol purple) with 15 g/L agar was used to isolate Enterococcus faecalis (previously, Streptococcus faecalis; Schleifer and Kilpper-Balz 1984) grown aerobically at 37 °C. ST (Streptococcus thermophilus) agar (10.0 g of tryptone, 10.0 g of sucrose, 5.0 g of yeast extract and 2.0 g K2HPO4 dissolved in 1000 mL distilled water) (Dave and Shah 1996) was used to isolate Streptococcus thermophilus, grown aerobically at 37 °C. Standard YPD media [1%
Table 1. Key characteristics of assessed probiotics.

| Probiotic | Labeled organisms | Species identified | Years expired | % Viability |
|-----------|-------------------|--------------------|--------------|------------|
| A         | Streptococcus salivarius | S. salivarius | 4            | 80.4       |
| B         | Lactobacillus paracasei ST11 | L. paracasei | 7            | 108.3      |
| C         | Lactobacillus acidophilus, Bifidobacterium lactis | L. acidophilus | 16           | 75.8       |
| D         | Lactobacillus casei, Streptococcus faecalis | L. casei, Enterococcus faecalis | 13           | 107.5 (L. casei), 100.4 (S. faecalis) |
| E         | Lactobacillus rhamnosus GG | NA | 17           | 0.0        |
| F         | L. acidophilus NAS super strain | NA | 15           | 0.0        |
| G         | L. casei HA-108, L. rhamnosus HA-111, Bifidobacterium breve HA-129, Bifidobacterium longum subsp. longum HA-135, L. acidophilus HA-122, Lactobacillus plantarum | L. casei, L. fermentum, L. rhamnosus | 6            | 87.8       |
| H         | L. acidophilus | NA | 22           | 0.0        |
| I         | L. rhamnosus | L. rhamnosus | 16           | 65.7       |
| J         | L. rhamnosus HA-111, L. casei HA-108, L. plantarum HA-119, L. acidophilus HA-122, B. bifidum HA-132, Bifidobacterium longum HA-135, L. fermentum HA-179, L. salivarius HA-181, Lactobacillus bulgaricus HA-137 | L. rhamnosus, L. fermentum | 6            | 92.9       |
| K         | L. plantarum | NA | 20           | 0.0        |
| L         | L. rhamnosus, Streptococcus thermophilus, Lactobacillus delbrueckii | NA | 10           | 0.0        |
| M         | L. acidophilus, L. casei, L. rhamnosus, B. longum, L. plantarum, L. fermentum, L. lactis, S. thermophilus, B. breve, B. bifidum | L. rhamnosus, L. lactis | 6            | 92.2 (S. thermophilus), 98.1 (other strains) |
| N         | L. reuteri ATCC55730 | NA | 15           | 0.0        |
| O         | L. paracasei (F-19), L. rhamnosus (HN001), L. acidophilus (LA-1), B. lactis (HN019), B. bifidum (TB-12) | NA | 10           | 0.0        |
| P         | L. acidophilus, B. lactis, B. bulgaricus, S. thermophilus | NA | 18           | 89.9       |
| Q         | L. rhamnosus Rosell-11, L. acidophilus Rosell-S2 | NA | 15           | 0.0        |
| R         | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 1            | 86.7       |
| S         | Saccharomyces boulardii | S. boulardii | 11           | 81.1       |
| T         | S. boulardii | S. boulardii | 13           | 98.3       |
| U         | S. boulardii | S. boulardii | 13           | 98.3       |
| V1        | L. reuteri RC14, L. rhamnosus GR1 | NA | 14           | 0.0        |
| V2        | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 11           | 95.9 (L. reuteri), 94.4 (L. rhamnosus) |
| V3        | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 8            | 98.4 (L. reuteri), 90.0 (L. rhamnosus) |
| V4        | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 6            | 91.9 (L. reuteri), 91.3 (L. rhamnosus) |
| V5        | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 11           | 90.6 (L. reuteri), 93.0 (L. rhamnosus) |
| W         | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 8            | 84.6 (L. reuteri), 85.5 (L. rhamnosus) |
| X         | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 13           | 101.0       |
| Y1        | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 12           | 99.1 (L. reuteri), 94.6 (L. rhamnosus) |
| Y2        | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 12           | 99.1 (L. reuteri), 94.6 (L. rhamnosus) |
| Z1        | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 13           | 92.7       |
| Z2        | L. reuteri RC14, L. rhamnosus GR1 | L. reuteri, L. rhamnosus | 13           | 100.8       |
| AA        | L. reuteri RC14, L. rhamnosus GR1 | NA | 11           | 0.0        |

NA: not available. % viability is the proportion of cells present at expiry that remained viable at testing, shown here after logarithmic transformation.

A—Dr. Milk Probiotic Milk, Blis Technologies Ltd, Otahu, New Zealand; B—DS, Innene, France, France; C—Bio-Tura, Pharma Nord, Mariton, New Jersey, United States; D—Yakult RI, Yakult Honsha Co., Tokyo, Japan; E—Culturrelle, CAG Functional Foods, Omaha, Nebraska, United States; F—GY-CY-TREN FLORA, Natren, Westlake Village, California, United States; G—Ultimate Multi Probiotic, Natural Factors, Canada; H—Milk-Free Acidophilus, Schiff Vitamins, Salt Lake City, Utah, United States; I—Florogyn vaginal capsule, Laboratories IPRAD, Paris, France; J—Ultimate Flower, Renew Life, Oakville, Ontario, Canada; K—Plantadophilus, Transformation Enzyme Corp., Houston, Texas, USA; L—Yogurt, Institut Rosell, Montreal, Quebec, Canada; M—Women’s Multi Probiotic, Natural Factors, Canada; N—Reuterin, BioGaia AB, Stockholm, Sweden; O—Theraflash, Therabiotics, Inc., Victoria, Minnesota, United States; P—Trevis, Chr. Hansen BioSystems A/S, Hørsholm, Tanska, Denmark; Q—Entero-Dophilus, Wilcox et al., USA; R—Investigational drug, Chr. Hansen BioSystems A/S, Hørsholm, Tanska, Denmark; S—Florastrant, MFI Pharma, Richmond Hill, Ontario, Canada; T—Lacticlan, EMS Sigma Pharma, Hortolândia, São Paulo, Brazil; U—Floratil 200, Merck, Kenilworth, New Jersey, United States; V—UREX-cap-S, manufactured by Chr. Hansen, Hørsholm, Denmark, received for clinical studies; W—RepHresh Pro-B, manufactured by Chr. Hansen, Hørsholm, Denmark, distributed by Lil’ Drug Store Products, Inc.; Cedar Rapids, IA; X—Bio-LiFE PRO-UTiX, manufactured by FA Herbs Sdn Bhd, Kuala Selangor, distributed by BiO-LiFE Marketing Sdn Bhd, Kuala Lumpur, Malaysia; Y1—2—Femophilus, manufactured by Chr. Hansen, Hørsholm, Denmark, distributed by Jarrow Formulas, Los Angeles, CA; Z1—2—Ecocflora, manufactured by Tablets (India) Ltd, Pondicherry, India; AA—Lacilbios feminia, manufactured by Exelit, Poland.
For probiotic products labeled as containing only one strain, DNA extraction and PCR amplification was used to isolate different lactic acid-producing bacterium, from the mixed culture (Lee and Lee 2008).

**RESULTS AND DISCUSSION**

For all probiotics, the proportion of cells that remained viable at the time of testing was compared with the time since expiry (Fig. 1B). There was a negative relationship between the time since expiration and viability, suggesting that after expiration, viability decreases over time. However, there is no specific point at which viability ceases.

The correlation between years expired and viability seen in this study is similar to several other studies of probiotics (Mary, Moschetto and Tailliez 1993; Teixeira et al. 1995; Borchers, Keen and Gershwin 2004; Champagne, Gardner and Roy 2005; Semyonov, Ramon and Shimoni 2011; Mizielińska et al. 2017). This result is expected because older probiotics have been exposed to external stresses (such as oxygen, light and moisture) known to cause microbial death (Tripathi and Giri 2014) for more extended periods, resulting in greater microbial death.

Pill type (encapsulated, compressed powder or loose powder) had no impact on the viability; most were encapsulated. Evaluation of the manufacturers’ probiotic labels was confirmed with either sequencing or selective media (Table 1). For viable

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Figure 1. Probiotic viability past expiration. (A) Expired probiotics with more than six remaining doses. (B) Graph of remaining viability (%) of expired probiotics versus years since expiration.

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Statistical analysis

The proportion of cells that remained viable at the time of testing was used as a measure of viability. Data were log-transformed to increase validity, additivity and linearity. All graphs were plotted and statistically analyzed using GraphPad Prism 8 (La Jolla, CA). Significantly higher viability in package type and packaging materials was identified using one-way analysis of variance (ANOVA) and Tukey’s honestly significant difference (HSD) post hoc test with Dunnett’s multiple comparison test. An unpaired t-test determined significantly higher viability based on storage conditions. One-way ANOVA with Tukey’s was used to analyze all other violin plots. Spearman correlation was used to analyze years vs viability.

For probiotic products containing multiple species that were differentiable by selective growth media, all differentiable colonies were collected. DNA was extracted from the samples using the InstaGene Matrix (Bio-Rad) following the manufacturer’s protocol, or the microwave method. For the microwave method, a small portion of a colony is spread on the inside of a PCR tube and then microwaved on high for 3 min and then PCR mix is added directly to this tube.

Bacterial DNA PCR protocol: 5 μL 10× PCR Buffer, 3 μL MgCl₂ (50 mM), 4 μL BSA (10 mg/μL), 2 μL pA (100 μM), 2 μL pH (100 μM), 1 μL Taq, 2 μL dNTP (10 μM), 2 μL template DNA and then topped off to 50 μL with NFH₂O. Run at 95 °C for 2 min, then 30 cycles at 95 °C for 30 s, 55 °C for 30 s and 72 °C for 1 min followed by a 10-min hold at 72 °C. PCR of bacterial sequences was conducted with 16S rRNA primers pA (AGAGTTTGATCCTGCGCTCAG) and pH (AAGGGGTGATCCAGGCCGGA) (Ulrike et al. 1989).

Fungal DNA PCR protocol was taken from White et al. (1989). PCR of fungal sequences was conducted using ITS primers, ITS1 (TCCTAGGTTGAACTTCGCGG) and ITS4 (TCTTGGCTATTGATAT GC) (White et al. 1989). Samples were amplified using Taq DNA polymerase. Reactions were primed at 95 °C for 5 min, followed by 35 cycles of 95 °C for 30 s, 50 °C for 30 s, 72 °C for 1 min; and a final extension at 72 °C for 10 min. Nucleotide Basic Local Alignment Search Tool (BLAST) was then used to determine the identity of the bacterial and yeast species in the probiotic.
probiotics, all consistently matched up to the manufacturers’ labeling. This study did not identify any microorganisms that were not listed on the product labels, although not all labeled microorganisms were identified. All products with more than 6 unit dosages are shown in Fig. 1A. These results indicate that labeling of genus and species in the products tested was accurate. This finding is interesting since past studies have shown inaccuracies in scientific labeling of probiotic genera and species (Weese 2003; Coeuret, Gueguen and Vernoux 2004). One evaluation of Lactobacillus probiotics found that 3 of the 10 human probiotics products had misidentified the species present (Coeuret, Gueguen and Vernoux 2004). A second study showed incorrect labeling of 9 out of 21 products, particularly in veterinary probiotic products (Weese 2003).

The impact on viability of the numbers of species or strains in a product was assessed, with between 1 and 12 present in a given product. There was no difference in viability between one or more species when looking at total cell counts of the products or individual species cell counts where tested (Fig. 2A). In this study, the number of species in a probiotic product had no observable impact on viability after expiration; however, only limited investigations of individual strain’s viability were conducted here.

When recommended storage conditions were not followed, there was a significant decrease in strain viability past expiration [Fig. 2B; P < 0.05 (P = 0.0191) unpaired t-test, n = 33]. One probiotic was stored in an ‘ultra-protective casing’ and it had 108% label claim viability at 7 years past expiration (Fig. 2C). In general, manufacturers recommend that probiotics be stored in a cold, dark, dry place or a refrigerator. Most of our expired probiotics were stored in a dark, dry box or a refrigerator for the time in our possession. This result is consistent with the literature, which indicates that refrigeration leads to higher viability than storage in ambient conditions (Weinbreck, Bodnár and Marco 2010; Tripathi and Giri 2014; Pimentel et al. 2015; Mizielińska et al. 2017). Overall, only two of the seven probiotics stored not according to the manufacturers’ instructions had viability. Probiotics A and B had much higher viability than the other incorrectly stored probiotics. This could be because both expired relatively recently (4 and 7 years ago, respectively) and were kept in metal packaging (Table 1).

Packaging material had a significant impact on bacterial viability, while capsule composition did not. The protective abilities of various packaging materials were evaluated (Fig. 2D and E). Capsule composition was categorized based on three main ingredients found in most of the tested probiotics (Fig. 2F). Probiotics formulated in capsules in a metal container were found to have significantly higher viability compared with a similar product in a plastic bottle [P < 0.05 (P = 0.0476) one-way ANOVA with Tukey’s, n = 33]. Based on packaging material alone, metal was also found to have significantly higher recoverable viability than plastic [P < 0.05 (0.0106) one-way ANOVA with Tukey’s, n = 33]. Although there is no significant difference between plastic and glass overall, smaller scale analyses revealed that there may be a difference in some instances. Probiotics T and U (Table 1) expired 13 years ago were stored correctly and presumably contained the
same organism (S. boulardii 17). Probiotic T, packaged in a plastic sachet, had no viability, while probiotic U, packaged in a glass bottle, had 98.3% viability. All other comparisons between packaging material and dosage composition were found to be non-significant. The type of seal used on the packing lids was not evaluated here and may have also been a factor.

Probiotics B, D, X and Z2 (Table 1) contained more than the manufacturers’ labeled microbial amount; at the point of testing, they had over 100% viability. The higher recoverable CFU than labeled may be due to ‘front loading’ of the product. Probiotics B and D were both contained in metal, while probiotics X and Z2 were in glass bottles with light protection. There was no other correlation between the probiotics; therefore, this difference may be due to packaging materials. Overall, glass and metal were found to be better packaging material than plastic to ensure the long-term viability of probiotics, though further studies with greater numbers are required to confirm this.

The results of this study suggest that packaging type may impact the long-term viability of probiotics after expiration. This is consistent with several other studies looking at the effect of packaging on probiotic viability prior to expiration (Talwalkar and Kailasapathy 2004; Chaikham 2015; Pimentel et al. 2015). This is attributed to the levels of protection against exposure to oxygen, light and moisture, which varied in each format (Mizielińska et al. 2017). In particular, plastic has a very high oxygen permeability compared with other materials (Mizielińska et al. 2017); so, the observed difference between metal and plastic packaging could be ascribed to their differing permeability to oxygen. Glass has been found to have a protective effect on probiotic viability (Weinbreck, Bodnár and Marco 2010; Tripathi and Giri 2014), though this was not observed in this study. We also need to consider that not all products in glass containers were stored according to the manufacturers’ instructions, and that these products had been exposed to stresses for longer than those considered in past studies. The lack of a significant difference between glass and plastic might also be because glass, despite providing better protection against oxygen, allows more light to reach the probiotic. Over time, all packages lose their integrity and allow contaminants that lead to microbial death, regardless of composition or format. These results show that none of the packaging tested consistently provided adequate protection over long periods of time.

This study has some limitations to be considered when interpreting the results. First, we used the proportion of cells remaining to quantify viability. This proportion was computed under the assumption that the number of live cells present at the time of expiry was exactly as guaranteed by the manufacturer. Though this was acceptable for the study, it is likely not accurate across all products because manufacturers add excess bacteria or yeast to ensure their products contain the guaranteed count at end of shelf life. Since the true count at the time of expiry may have been greater or less than estimated, this could have increased or decreased the proportions used in our analysis. In future experiments, it may be better to quantify the number of dead cells and use this to calculate a proportion that more accurately represents the change in viability. Second, the groups considered in each analysis varied in terms of characteristics other than the ones used to define the collection (e.g. the products in the ‘stored correctly’ group varied in terms of time since expiry, packaging and the number of species). Ideally, the groups would differ only in terms of the variable being investigated, so that any conclusion can be clearly linked to that factor. Third, many of the products were stored according to the manufacturer’s instructions that may not be representative of the general public. It would be beneficial to do a larger study looking into extending the expiration of probiotic products and a survey of storage conditions in further research.

CONCLUSION

This study found that many probiotic products retained their viability long after expiration, albeit lower than the recommended threshold for efficacy. Currently, expired probiotics are not safe to consume past expiration despite containing viable cells due to the lack of safety studies. Companies often calculate expiration date based on accelerated studies alone. Ideally, they should re-evaluate their viable count to determine expiration in real time to more accurately reflect when the product no longer meets its efficacy threshold.

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SUPPLEMENTARY DATA

Supplementary data are available at FEMSMC online.

Conflict of interest. None declared.

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