

PROBIOTICS (Comment)

Has the time come to start using probiotics more widely?

Much research has been done into the use of probiotics in children in the last few years, and a wide range of products have been studied. Some trials have looked into the use of a single organism (usually one of the bifidobacteria) although, on examination, what was actually being given was often really more complex than that (Marcobal *et al.*, 2008). Other trials have used mixtures and this, as we know, mimics more closely what is going on in the healthy human gut. Cochrane Reviews currently show that use can only do so much to reduce allergic disease (Osborn and Sinn, 2007) or to make eczema less troublesome (Boyle *et al.*, 2008), but may reduce the severity of infectious diarrhoea (Allen *et al.*, 2003) and make the diarrhoea that is sometimes triggered by antibiotic treatment less common (Johnston *et al.*, 2007). The main focus of early use in the preterm baby has, however, been the potential for treatment to reduce the risk of necrotising enterocolitis (NEC) and this has been the focus of yet another Cochrane Review (Al Faleh and Bassler, 2008).

There have, in fact, been three systematic reviews of this issue (Barclay *et al.*, 2007; Deshpande *et al.*, 2007; AlFaleh and Bassler, 2008). Although the review by Al Faleh was the last to appear it seems, at the moment, to be the least complete, but it is currently being updated. Deshpande and his colleagues have, however, already updated the review they originally published in the *Lancet* in May 2007, in order to include the new trial involving 434 babies weighing less than 1500g at birth that was published by Lin *et al.* in 2008, and the slightly smaller trial involving 186 babies that was published by Samanta *et al.* in 2009. This update, which was presented at the meeting of the Perinatal Society of Australia and New Zealand in Darwin in April 2009 (Deshpande *et al.*, 2009), further strengthens the conclusion that had already been reached by the three earlier reviews – that use can more than halve the risk of NEC serious enough to require surgery, and the risk of post-entry death. Full enteral feeding seems to be achieved significantly sooner, and there is no evidence of any increase in the risk of sepsis. In the most recent meta-analysis data from 9 trials involving more than 2000 babies found that babies weighing less than 1500g at birth who were offered probiotics from birth were at much less risk of developing NEC (Bell classification stage 2 or greater) – relative risk 0.34 (95% CI 0.20 to 0.54; $P > 0.00001$). Death was also less common – relative risk 0.41 (95% CI 0.28 to 0.61) $P > 0.00001$. The risk of sepsis in the two trial groups was very similar (RR 0.97 [95% CI 0.80 to 1.19]), but full enteral feeding was, on average, achieved 5 days earlier in the babies given a probiotic, with their feeds, once or twice a day.

NEC is a potentially devastating condition in which patchy areas of bowel wall are invaded by gas-forming organisms and quickly become necrotic and, almost fifty years after the condition was first recognised, surgery still remains the only effective form of treatment (Lin and Stoll, 2006). Once classic NEC is distinguished from spontaneous focal intestinal perforation (which is also common in babies of less than 28 weeks gestation) it becomes clear that true NEC currently affects some 6% of all babies as immature as this, and that half of these become ill enough to require surgery (Gordon *et al.*, 2007). As many as a third of these will die, while quite a lot of the survivors will be left with a damagingly short gut (Cole *et al.*, 2008). With no new approach to treatment in sight, prevention has to be the main focus of attention.

It is now accepted that feeding with breast milk reduces the risk of NEC (McGuire and Anthony, 2003; Hanson *et al.*, 2003), and the implication is that there must be some role that only human milk can play in facilitating the development of a secure mucosal barrier, and a healthy balance of bacteria in the intestinal tract (Grönlund *et al.*, 1999; 2000; Hooper, 2004). Indeed we already know that the microflora in the gut of the very preterm baby lacks the bio-diversity that is seen within days in a healthy baby born at term, and that this is particularly true of preterm babies who are not given breast milk (Grewolb *et al.*, 1999). We have also known for 15 years that prophylaxis with an unabsorbed oral antibiotic can reduce the risk of serious NEC, (Bury and Tudehope, 2001) although this approach has never been taken up because of untested fears that it might increase the number of multiply resistant, dangerously pathogenic, bacteria.

Epidemiological studies strongly suggest that babies who are not given at least some milk fairly soon after birth becomes progressively more vulnerable to late-onset (nosocomial) infection (Stoll *et al.*, 2002; Flidel-Rimon, *et al.*, 2004), although it is not clear whether this is because milk delivers some protective benefit or whether it is because these babies stop having long lines in place and stop being given parenteral nutrition much sooner. While there is, conversely, some suggestion that trying to build up enteral intake too rapidly may make serious disease slightly more likely (Henderson *et al.*, 2009) the evidence for this is still very weak (McGuire and Bombell 2008; Bombell and McGuire, 2008) and a 'gradualist' approach risks further lengthening the period of arrested post-delivery growth that almost certainly accounts for at least some of the long term cognitive deficit with which almost all babies born this early currently have to contend in later life.

Given what we now know about the time it takes for the gut of the preterm baby to become colonised by the wide range of organisms that contribute to healthy gut function (Claud and Allen, 2008) early prophylactic treatment with an oral probiotic seems a very logical strategy. The fact that very low birth weight babies who are given antibiotics empirically for longer than average in the absence of any evidence of overt infection are substantially more likely to develop NEC and to die (Cotten *et al.*, 2009), also points to the importance of

helping these babies to acquire, right from the outset, a healthy balance of gut bacteria. Systemic *Candida* infection is a worryingly common problem in the very preterm baby and, in the only trial that seems to have looked at this as yet, probiotic use reduced the number of babies whose gut became colonised with *Candida* species and the intensity with which it became colonised (Manzoni *et al.*, 2006).

All this makes it difficult to understand why it is taking clinicians so long to start using early exposure to a mixture of probiotic organisms to facilitate balanced colonisation, especially in babies of less than 32 weeks gestation – the babies at highest risk of developing NEC. Observational studies have been pointing to the benefit to be derived from doing this for ten years (Hoyos, 1999; Lian-Qiao *et al.*, 2006; Satoh *et al.*, 2007), and formal clinical trials have also been showing increasingly powerful evidence of efficacy for more than four years (Dani *et al.*, 2002; Costalos *et al.*, 2003; Bin-Nun *et al.*, 2005; Lin *et al.*, 2005). Neither is there any evidence of heterogeneity in the findings to suggest that the finding of any meta-analysis needs to be interpreted with caution (Egger *et al.*, 1997).

The reasons for hesitancy seem to be various and complex. Some clinicians feel that this is a topic about which they are, at the moment, rather poorly informed. Uncertainty on the part of the regulatory authorities over whether to classify probiotics as a food or a medicine has caused confusion and seriously delayed research in some countries (Hibberd and Davidson, 2006). Treating them as a foodstuff will generally lead to health claims but treating them as a medicinal product must necessarily involve therapeutic claims, and the regulatory implications are very different. Many clinicians also worry about the reliability of some of the products currently on the market, doubt whether anybody is responsible for quality control, and remain very unclear as to who needs to take ultimate responsibility for initiating use even though the products used in several of the largest trials have long been on sale to the general public.

Many also worry that, although several trials have now shown short term benefit, few have yet included further follow-up after discharge in order to document the longer term outcome. There is, however, one reassuring abstract reporting the two year term outcome for 83% of the 367 babies involved in the first of the two recent trials undertaken in Taiwan (Lin *et al.*, 2008). A definitive paper reporting these outcomes should appear in print next year, and similar information will eventually become available from the 434 children in the second study. In addition, several quite large trials of long term postnatal use have been undertaken in the last few years. One trial which recruited 284 babies, gave half the babies a probiotic in the belief that this might make diarrhoea less likely, and monitored progress for a year to confirm that such treatment was both safe and well tolerated (Chouraqi *et al.*, 2008). It was. Another trial recruited 925 babies judged to be at high risk of allergy, and followed them for two years (Kukkonen *et al.*, 2008). This also found treatment safe and well tolerated. A third trial recruited 171 women with a strong history of atopy who planned to breast feed and undertook a double-blind placebo-controlled trial with nutritional modulation by dietary counselling and sustained pre- and post-delivery maternal probiotic supplementation. Probiotic use had a protective effect against sensitisation (as assessed using skin prick tests) especially in infants at high hereditary risk due to maternal sensitisation (Huurre *et al.*, 2008). The children were followed for a year and no adverse effects were reported. A sceptic would point out that these three trials involved term babies rather than preterm babies, but the report from Taiwan related to babies who weighed less than 1500g at birth, and the neurodevelopmental outcomes for preterm babies in observational studies have been equally reassuring (Schulzke *et al.*, 2007),

There is, of course, a difference between giving a probiotic to a gut that has already been colonised by a wide variety of organisms, and giving it to a gut that is still almost sterile. Some may consider this risky and 'unnatural'. Others may think that paying more attention to early priming could be the key to success. Few realise that several aspects of the way we currently manage birth can have a major impact on the way the gut does initially become colonised. Babies normally acquire a range of organisms from their mothers during birth, but Caesarean delivery can, on its own, be enough to have an effect on the flora in the child's gut that is still detectable seven years later (Salminen *et al.*, 2004). Peripartum antibiotic treatment may also have delayed consequences for the infant (Bedford-Russell and Murch, 2006). The prolonged separation, early antibiotic exposure and delayed feeding that occurs to every very preterm baby cannot but have an even more profound impact, especially during the first few weeks of life. Perhaps we ought to be exerting an active influence on the way the gut becomes colonised in these babies, instead of leaving it to chance, as happens, by default, at the moment.

The three recent systematic reviews have all said that further trials are necessary, and that much still remains to be learnt about when to start treatment, how long to continue treatment, the best product to use, and the best dose to give. Intuitively however it would seem logical to start treatment early, but only continue treatment for more than a month if antibiotics are still in use. And if treatment is as safe as it seems to be, the dose given is probably not critical as long as it can be shown to have achieved, and sustained, the desired effect on the balance of bacteria in the gut. And, in saying that "further trials are necessary", the reviews were not necessarily implying that there is not, as yet, enough evidence to advocate some use – they were merely saying that there is more still to learn about the best way to optimise use. Indeed the number of further trials

needed might be smaller than we think if it proved possible to merge the raw data from every child recruited into all the various trials that have already been done, instead of simply relying, as now, on summary findings.

There are those who currently argue that it is possible that probiotic treatment is only necessary, or only works, in formula fed babies, forgetful of the fact that *all* the babies recruited into the two largest trials were initially fed breast milk and that only a third received any formula milk during the first six weeks of life.. Others are concerned that, although probiotics seems to work for children of one ethnic group, or experiencing one particular tradition of nursing care, it might not work for those from a very different ethnic group, or nursed in a rather different way. That rather overlooks the fact that trials have already been done Italy, Greece and Israel as well Taiwan, Japan and India. The 'generalisability' of any trial finding is always an important issue, but there comes a point where that can not, on its own, be used to justify the mounting of further trials when a more detailed interrogation of all the trial data that is already available, if pooled, might well serve to answer these questions without requiring any more placebo-controlled trials or delaying the much more general adoption of a very promising strategy for reducing the current scourge of NEC.

Others point to the fact that, while there is now evidence of benefit for all the babies weighing under 1500 grams recruited into the various trials to date, it is less conclusive for those in the trials who weighed less than 1000g. There are, however, dangers in taking too much notice of sub-group analyses, especially if they were not prespecified before the study began. Many will recall the notorious conclusion that many derived from the first American study of the use of dexamethasone antenatally to speed fetal lung maturation (Collaborative Group on Antenatal Steroid Therapy, 1981). The perception was that this only offered benefit to the female babies of black mothers (Roberton, 1982) – an interpretation of the trial data that set back the more general adoption of one of the most important advances in neonatal care of the last half century for almost ten years. While there can be good reason to look with particular care for possible adverse effects of early probiotic treatment in the smallest and most vulnerable babies, there is no good theoretical reason for supposing that something that seems to offer clear benefit to babies weighing 1.0–1.5 kg at birth does not have the potential to offer similar protection to babies weighing less than this at birth.

Will clinicians look back in ten years and wonder why it took so long for most to adopt a strategy that is simple and cheap, and seems, after a decade of study, to have no detectable adverse consequences, when it has already been shown in nine randomised controlled trials to more than halve death from NEC in babies weighing less than 1500 g at birth? NEC is, after all, by far the commonest reason why very preterm babies who have survived the first week of life unscathed currently fail to go home alive. Is the “herd” instinct at work here? The advantages of any new strategy often become obvious before the disadvantages, so there is a strong case for telling parents that the strategy is relatively new and for only using it at the moment with parental consent. Such an approach would soon make it clear what parents think we should be doing given our current level of knowledge. Do we, knowing what we now know, have the right to deny parents the option of giving a probiotic if that is what they would like? While early use may not really be capable of reducing the risk of NEC quite as much as evidence currently suggests, there is almost no evidence that use ever does any harm.

And the robust evidence of efficacy now available should not mean that the trials that are currently recruiting need to stop because, as the answer to one question becomes clear, half a dozen other important questions immediately surface. The problem is that the rigid way that most trials are now regulated makes such flexibility difficult to achieve. The time it currently takes to plan, finance, approve, recruit and report a trial of sufficient size to get a reliable answer to even a single question is not keeping pace with the speed with which knowledge is advancing. This needs to change. However rising use does not need to bring research to a stop.

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