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Clinical trials to prevent/treat childhood asthma: methodologic issues

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Keywords: asthma • clinical trials • primary prevention • vitamin D

There is currently much excitement about clinical trials for the primary and secondary prevention of childhood asthma [1-3]. For the first time in over 40 years there are real novel therapeutic possibilities that deserve testing. In thinking about the design of these trials, it is critical to consider four issues: the traditional validity threats to clinical trials, how fetal lung development influences clinical trial design, how asthma natural history and asthma subphenotypes influence trial design and finally, drug dosing for these studies. As might be suspected, fetal lung development has its greatest impact on primary prevention trials and asthma sub phenotypes have its greatest effect on secondary prevention trials.

Although people often focus on the randomized controlled nature of clinical trials, the methodological issues of blinding and randomization address internal validity, and not the source population from which trial subjects are drawn. The greatest validity threat to any clinical trial is the external validity generalizability of results, can the trial results be extrapolated to the population at large and what will be the population impact of the new treatment or preventative.

Concrete examples of these generalizability issues can be seen in the source population for two of the new primary prevention trials in childhood asthma: the Vitamin D Antenatal Asthma Reduction Trial (VDAART) and the recent trial of vitamin C to prevent asthma in smoking pregnant women [1,2]. In the former case, VDAART is focused on women with a prior personal, or family, history of asthma, or allergies and giving them vitamin D to prevent asthma in their offspring. If successful, it will be a generalizability leap to all pregnant women, even those with a low genetic risk of asthma in their children, to extrapolate the trial results. In the latter case of vitamin C, the source, or target population, is the 12% of pregnant women who smoke and, as yet, there is no evidence that vitamin C will prevent asthma in nonsmoking pregnant women. This issue of generalizability is important in targeting public health interventions particularly in the area of primary prevention.

Primary prevention trials in childhood asthma

Previous primary prevention trials in asthma focused on minimizing exposure to allergens [4]. More recently, the use of dietary supplements in pregnancy has received attention, and this editorial will focus on these. This is an extension of the work of David Barker who initially hypothesized that maternal diet during pregnancy was an important driver for programming of fetal development [5]. Fetal lung development is a complex process that is controlled by poorly understood epigenetic and genetic mechanisms. Most importantly, the process begins very early in pregnancy with lung budding in the fourth week of gestation, proceeding through the development of the airways early in the second trimester (10-18 weeks) and alveolarization in the third trimester (18-40 weeks) [6]. Given this clear-cut developmental progression, the scientific question becomes, when to intervene? It would be ideal to begin all



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primary asthma prevention trials prior to conception to allow for maximal effect on the developing fetal lung. Indeed vitamin D may have effects on fertility and implantation of the fertilized egg that would suggest biologic reasons why such an approach would be best [7]. However current trials have accepted that such an approach may not be practical initially as investigators begin to get these novel studies off the ground.

It is important to consider the impact on trial results of misjudging the timing of when to start primary prevention treatments. Certainly starting later in pregnancy will potentially result in a null bias and could lead to completely negative results when the treatment under study might actually be effective if it were started earlier in fetal lung development. VDAART began enrollment at the 10-18th week mean week 14, while the vitamin C trial enrolled mothers a bit later, but had primate data suggesting that treatment in that developmental window would be effective [2]. There are genomic data to suggest that vitamin D is involved very early in lung development [8], so if VDAART shows an effect on asthma prevention, the effect might be even greater if the treatment were started earlier.

With regard to primary prevention trials, the issue is less what subtypes of asthma to focus on, and more having a clear understanding of asthma natural history. In total, 40% of children wheeze with a viral illness in the first 2 years of life. This wheezing with infections decreases to 20% by age 3 but it takes until age 6 to definitively say that a child has asthma based on both symptoms, allergy testing and lung function assessment. Thus, trials that only last until the end of the first year of life will not be able to definitively say whether there is an effect on asthma prevention until the children are followed up much later in life, usually after age 6. This does not preclude the value of shorter trials, like the vitamin C trial, that utilized pulmonary function and other intermediate outcomes as end points, but it should be emphasized that definitive diagnosis of asthma is not possible in very young children and the certainty of the diagnosis increases with increasing age of the child. Compromises will likely have to be made to control both the length of these trials and their cost.

Another very important issue in these early life primary prevention trials is whether the treatment of the mother during fetal development is sufficient to prevent disease or whether it is also necessary to treat the child as well as the mother. When designing the VDAART trial we initially wanted to do a cross over study at birth so the children could be re-randomized to treatment with vitamin D or not. This would have given us four groups with mother' and their off spring who were both treated and untreated. This factorial design was optimal but was abandoned due to the prohibitive costs. We opted instead to follow the trial participants until age 6 to see how long the effects of maternal supplementation alone would persist.

The final issue in the area of primary prevention is what nutrients should be tested and at what dose. The most interesting candidates that have come from both animal studies and human observational work are: retinoic acid (vitamin A), folate, other B vitamins (choline betain), vitamin C, vitamin D, n-3 fatty acids and cod liver oil (vitamin A, vitamin D and n-3 fatty acids).

This list can be parsed in a variety of ways, but clearly vitamin D is the most attractive candidate. First, 70% of all pregnant women are deficient so there is actual evidence of deficiency in western-developed populations [9]. Second, the effects on fetal lung and immune system development are there and the epidemiologic data are compelling [10]. Finally, dosing has been well worked out with studies in pregnant women defining a dose of 4000–6000 IU of vitamin D daily as leading to a serum level of at least 30 ng/ml, the minimum immune sufficient dose, with no side effects [11].

Retinoic acid and folate are likely to be interesting primarily as to whether they might interact with vitamin D and other vitamins. Retinoic acid and vitamin D share, and compete for, the same receptor and the effects of each vitamin alone on fetal lung development is different in the absence of the other and maximal if both are present [12]. Despite known developmental influences, a primary deficiency of retinoic acid and folate are unlikely, as maternal diets are supplemented in western-developed countries for both of these vitamins so the issue is primarily interaction with vitamin D, and variation within the range of normal, rather than deficiency. We know that retinoic acid can be supplemented safely at the dose seen in cod liver oil as that treatment was used for at least a century without complication [13]. Folate raises the question of other methyl donors that are part of the B vitamin pathway, and lesser-known B vitamins such as choline and betain may be of relevance [14]. Again folate is already within the range of normal in the current maternal diet so any effect on asthma is likely due to interaction with another nutrient or variation within the range of normal. Prior to 1946, when pregnant women and their offspring took cod liver oil, the interaction of omega 3 fatty acids and vitamin D was moot, but now that cod liver oil is no longer used, this scientific question should be revisited. While vitamin E seems to be less important in observational studies than vitamin D, it too deserves further study [15]. Finally, the recent successful completion of the use of vitamin C in pregnant smoking women to reverse

the effect of in utero smoke exposure begs the question of how vitamin C is working to prevent these adverse effects and again, the interaction with other nutrients is of research interest [2].

Secondary prevention trials

Unlike primary prevention studies of childhood asthma, with secondary prevention studies the major question is not the timing of the trial, but the phenotype of childhood asthma. It is critical to know the natural history of early life childhood asthma especially the finding that the majority of these subjects will get better with increasing age. There are three groups of wheezing children in early (0-3 years) childhood: transient wheezers, persistent wheezers and nonwheezers. Prior to age 5-6 it can be difficult to separate these three groups and it is only in retrospect that that can be done. Thus, trials in the 0-5 year age group will, of necessity, mix these three phenotypic groups. By age 5–6 childhood asthma can be definitively identified and can usually be grouped into subjects who will remit on its own as the child grows and those with more persistent disease. Persistent childhood asthmatics, usually atopic, with persistent symptoms, make up 10-20% of all childhood asthma and are the group most likely to have reduced growth in lung function growth, a greater number of exacerbations and will likely require inhaled steroids. It is this group that forms the group of greatest clinical concern and the one in whom clinical trials of novel treatments should be directed. Some of these subjects can be identified as early as age 3 years as they are repeatedly being hospitalized and require steroids to control their symptoms and may even have breakthrough symptoms/exacerbations on steroids.

What are the key secondary prevention trials that need to be done? At present there are *in vitro* data to suggest that vitamin D upregulates IL-10 at the airway epithelium and also enhances the effect of steroid and reverses steroid resistance [16,17]. This finding coupled with retrospective clinical trials data, and the high prevalence of vitamin D deficiency suggests that clinical trials of vitamin D with inhaled steroid make sense for the secondary prevention of childhood asthma [18–20].

The recent VIDA trial in adults looking at vitamin D to prevent exacerbations brings up some interesting study design issues [3]. Asthma Net decided that instead of performing a trial in children, where the literature suggested they would get the biggest effect, they chose to study adults with more severe and long-standing disease. In adults, supplementation for over 28 weeks, which was the duration of the trial, may be needed. This may have affected the results of

their primary outcome, which was time to first asthma treatment failure (defined in the trial as a composite outcome of decline in lung function and increased use of β-agonists, systemic corticosteroids, and healthcare). While the addition of vitamin D was found to decrease the overall asthma treatment failure rate (aHR = 0.8; 95% CI = 0.6-1.1) and overall asthma exacerbation rate (aHR = 0.63; 95% CI = 0.39-1.1), these did not reach statistical significance, likely due to the fact that there was a lower than expected event rate in the control group leading to inadequate power in the study. The addition of vitamin D did reduce the dose of inhaled steroids in the intervention group (p = 0.03) [3]. Therefore, an adequately designed and adequately powered clinical trial in children is needed. Admittedly, dosing is an issue but 4000 IU should work perfectly in children over the age of 3 years. Alternatively one could dose with two tablespoons of cod liver oil or its pill equivalent and avoid any problem with safety concerns. While there are other agents and nutrients to try for secondary prevention the overwhelming retrospective data and the low cost of the treatment make such a study a high priority.

Conclusion

This is a very exciting time to be doing asthma clinical trials and despite the complex methodological issues there is real promise for disease prevention and treatment. With regard to primary prevention of asthma, there has been recent focus on vitamins and nutrients, with the realization that intervention in the prenatal period is important. Many issues face these trials including the timing of the initiation of the intervention and the duration of the intervention. These issues need to be balanced with the real-world issues of cost and practicality in designing these trials. In contrast, the main issues facing secondary prevention trials are the heterogeneity of the asthma phenotype and the natural history of early wheezing in children. These are not insurmountable issues, but their careful consideration in the design of such trials will lead to better ability to prevent the consequences of childhood asthma.

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