Newborn Screening by Tandem Mass Spectrometry: A New Era

Soon after Guthrie (1) expanded newborn screening by adding galactosemia, maple syrup urine disease (MSUD), and homocystinuria to the original screening for phenylketonuria (PKU), he realized that screening would be more efficient and comprehensive if a single assay could be used to detect several disorders rather than the system of a separate bacterial assay for each disorder that he had developed. He tried many ways to make such a single assay—using multiple inhibitors and different strains of bacteria—but nothing worked, so he abandoned the idea. Others had the same idea but used chromatography rather than bacterial assays (2, 3). Unfortunately, paper chromatography was insufficiently sensitive for screening newborn blood within the first days of life when the specimen is collected. To compensate for this shortcoming and to further expand the coverage of disorders, paper or thin-layer chromatography has been used for screening newborn urine (4). But here again, chromatography has had substantial disadvantages. First, an additional specimen is required because urine cannot replace blood in detecting either PKU or congenital hypothyroidism, the two indispensable disorders in screening. Second, the urine specimen must be collected by a parent or physician after nursery discharge, introducing a logistical problem. Third, urine varies widely in concentration, producing many false-positive results in the more highly concentrated specimens. This leads to otherwise unnecessary, anxiety-provoking requests for repeat specimens. Conversely, false-negative findings may result from dilute specimens. Finally, many of the disorders identified in urine, such as histidinemia, iminoglycinuria, and Hartnup disorder, are benign (4). Consequently, newborn urine screening based on chromatography has been discontinued in two of the three places in which it was introduced, Australia (5) and Massachusetts, remaining only in Quebec (6). Thus, screening programs continue to rely on the “one test-one disorder” system.

Technology now allows a “sea change” in newborn screening. Tandem mass spectrometry (MS/MS) is the most important development in newborn screening since the addition of screening for congenital hypothyroidism in the mid-1970s. There are several reasons for its importance. It greatly expands newborn screening coverage of the metabolic disorders. In addition to PKU (7), it can identify at least 10 other amino acid disorders, including two (maple syrup disease and homocystinuria) (8, 9) that can be included in the current screening but increasingly are excluded because each disorder requires a separate bacterial assay. The expansion also extends to important disorders of organic acid degradation and fatty acid oxidation (10, 11). These 20–25 disorders are screened in the blood specimen, avoiding the need for an additional specimen. Only a single test is required, changing the screening system from “one test-one disorder” to the “one test-many disorders” concept that, whenever possible, should be a cardinal principle of screening. With MS/MS screening, programs need not discontinue or exclude screening for MSUD or homocystinuria on the grounds that additional tests for such rare disorders cannot be justified. Only one or two small disks (blood spots) from the specimen are needed for MS/MS, saving most of this increasingly valuable specimen for still other tests. Very importantly, the false-positive rate is lowered, despite a marked increase in the number of disorders covered. Neo Gen Screening of Pittsburgh, the program that has pioneered the application of MS/MS to routine newborn screening, has had a false-positive rate of only 0.26% (E.W. Naylor and D.H. Chace, personal communication). In comparison, the New England Newborn Screening Program, using traditional bacterial assay methodology, has had a false-positive rate of 1.5% (H. Levy, unpublished results) while covering substantially fewer disorders and identifying far fewer infants with metabolic disease.

The reduction of false-positive results is presented dramatically in this issue of Clinical Chemistry. Chace et al. (12) describe the use of MS/MS to re-test stored newborn specimens collected at early discharge (<24 h of age) from California infants in whom screening by fluorometric assay revealed PKU or variant PKU or who were classified as false positive. The phenylalanine concentration and the phenylalanine:tyrosine molar ratio determined by MS/MS identified all of the affected infants and eliminated 90 of the 91 false-positive results (12). Thus, MS/MS screening reduces the false-positive rate because it identifies disorders on the basis of improved quantitation via the stable isotope dilution technique together with the pattern of metabolite abnormalities as opposed to screening for a single metabolite increase that is often transient. The importance of reducing false-positive results in newborn screening cannot be overstated. False-positive results account for much of the expense of newborn screening and most of the anxiety. Follow-up for each such result requires searching for and re-testing the initial specimen, contacting the physician or clinic by phone call and/or letter for a second specimen, tracking receipt of the requested specimen, testing this additional specimen, and then notifying the physician or clinic of the second result (13). Any physician working in a newborn screening program can attest to the enormous anxiety this produces in families and the additional work required of the attending physician. During my 30 years in the Massachusetts and then New England Newborn Screening Program, fully 75% or more of my time was devoted to handling false-positive results.

If MS/MS technology is such an improvement over presently used methods, with fewer false-positive results and less expense over time because of the greater efficiency, why are newborn screening programs not rushing to adopt it? The wrong answer is that the technology is too new or unknown. It has been discussed widely in newborn screening circles and within the academic met-
able community. Two years ago, Clinical Chemistry published a comprehensive editorial by Sweetman (14) describing the importance of MS/MS for newborn screening. The correct answer requires an understanding of how newborn screening is conducted in the United States and most of the rest of the world. Screening requirements and the determination of which organizations may provide screening services are almost completely controlled by state or other governmental health departments. In most cases, these political entities have determined that they alone are qualified to provide newborn screening. However, government agencies are generally not distinguished by their technological innovation or by their readiness to incorporate new ideas. Their virtual dictum is “if it ain’t broke, don’t fix it.” In one sense, newborn screening “ain’t broke.” Almost all infants with PKU and congenital hypothyroidism are identified. Infants with three to five other disorders are also usually identified, at least in some programs. In another sense, however, it is “broke”. Many infants with disorders not covered by traditional newborn screening are not identified presymptomatically and become mentally retarded, develop “cerebral palsy”, or die suddenly. These infants and children, however, are followed in medical centers outside the purview of health department officials. In the United States there is often little communication between medical centers and newborn screening programs. Consequently, those who have knowledge of and expertise in these disorders have no authority in newborn screening, and those who have this authority are usually insufficiently informed and resistant to change—especially when they could be impacted personally by this change. Also inhibiting change is the complexity of MS/MS. With few exceptions, health department screening programs do not have and are unlikely to be able to acquire and retain the expertise required to adequately perform MS/MS analyses and interpret the results in a timely manner.

The solution may be a joining of both worlds. Health departments should form alliances with newborn screening programs experienced in implementing state-of-the-art technologies. The objection that private screening programs—even those dedicated to newborn screening and with extensive experience with advanced technology and a proven performance record in newborn screening—are not as good or as responsible as a public program is clearly not valid. In fact, many state newborn screening programs are very similar to private laboratories in that they charge for services, pay employees from these receipts, and realize a profit. Nevertheless, government health agencies have an obligation to determine the disorders for which mandatory screening is required and to establish the quality standards for organizations providing these services. The solution, therefore, is to develop a dual approach. Those state programs that wish to maintain strict control over the specimen itself should develop partnerships with programs having the expertise or should contract for newborn screening MS/MS services with such programs. Other states could allow hospitals to send the newborn specimen to any program dedicated to newborn screening that complies with regulations established by the government health agency. Out of this can emerge a restructuring of newborn screening, with a regional rather than the currently outmoded and inefficient state-by-state approach, and a national consensus on screening so that all infants and their families can receive the full benefits possible from one of the most important preventive approaches in medicine.

References


Harvey L. Levy

Children’s Hospital
300 Longwood Avenue, IC-106
Boston, MA 02115
Fax 617-730-0461