A HealthTech Historical Profile

Lateral-Flow, Point-of-Care Diagnostic Tests for Infectious Diseases

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MAIL
PO Box 900922
Seattle, WA 98109 USA

STREET
2201 Westlake Avenue, Suite 200
Seattle, WA 98121 USA

Tel: 206.285.3500
Fax: 206.285.6619
www.path.org

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PATH A catalyst for global health

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HealthTech Historical Profile: Lateral-Flow, Point-of-Care Diagnostic Tests for Infectious Diseases

The Problem

Very few primary health care facilities in the developing world are within reach of well-equipped and staffed clinical laboratories. Diagnosis of disease at the community level requires access at the point of care to rapid, low-cost, easy-to-use, and appropriate diagnostic tests for infectious diseases.

For example, more than 2 billion people live in malaria endemic regions of the world. As a result, more than 300 million new cases of malaria occur each year, resulting in several million deaths worldwide. But microscopy, the gold standard for diagnosing malaria, is only available and performed well at well-equipped laboratories with trained staff. A simple, rapid test has therefore been a high priority for development since such a tool available at the point of care would ensure that patients could receive proper treatment and that infection by other pathogens producing similar symptoms of acute fever would be excluded.

Similarly, syphilis, a sexually transmitted infection (STI) caused by *Treponema pallidum,* causes genital ulcer disease (GUD) and can easily be treated with penicillin, a relatively inexpensive antibiotic therapy. However, the ulcers produced by *T. pallidum* cannot dependably be distinguished in the clinic from other causative agents of GUD (e.g., *Haemophilis ducrey*, and Herpes Simplex Virus 2), and after early ulcerative infection, many cases are asymptomatic. If syphilis remains untreated, it may eventually cause long-term debilitating effects that are potentially fatal. Pregnant women who are infected can pass the disease to their newborn infants, where it can produce systemic infection and death. Syphilis also increases the risk of contracting or transmitting HIV and other STIs. Syphilis is generally diagnosed by an agglutination test requiring a centrifuge and rotator and performed in a laboratory. As a result, syphilis remained under-diagnosed and under-treated in most of the developing world where access to testing was limited. Even as late as 1996, no affordable rapid field test for syphilis was available for developing-world use.

Likewise, despite long-standing, global, public health efforts for their control, *Chlamydia trachomatis* and *Neisseria gonorrhoeae* infections still occur in epidemic proportions in the developing world. In 2000 WHO estimated there were 333 million new STIs yearly. For more effective control, STI-control programs need to provide for early and accurate diagnosis of symptomatic infection and identification of invasive, complicated, or asymptomatic infections. Control of STI is also considered to be an essential component in control of HIV/AIDS transmission. Available tests are expensive and/or technically complex, culture diagnosis is unavailable or prohibitively expensive, diseases are poorly recognized or asymptomatic, and syndromic management algorithms for women are neither sensitive nor specific. Simple, rapid tests for STIs that can be used at the point of care are urgently needed

Tuberculosis (TB), a bacterial disease caused by *Mycobacterium tuberculosis*, is a disease that has reemerged as a major health threat in the developed world and continues to produce significant morbidity and mortality world-wide. Globally, as many as 3 to 4 million deaths can be attributed to TB each year. These estimates are expected to rise dramatically given that TB is a common opportunistic infection for HIV-infected individuals at late stages of disease progression. A top priority for TB control programs is active case detection, confirmation of infection, and cure of all infectious cases in both low-and high-prevalence areas. Diagnosis of TB is difficult, however. Available diagnostic tools are seriously limited by their speed, cost, technical complexity, difficulty in scale-up, and less-than-ideal accuracy. A few key tools or interventions, if developed, could therefore greatly assist existing control programs.

Three-quarters of the world's population live in highly endemic hepatitis B virus (HBV) regions. Among the estimated 1 billion persons infected world wide, there are 300 million hepatitis B surface antigen (HBsAg) carriers and 2 million annual fatalities resulting from HBV infection. HBsAg carriers can be extremely infectious and therefore must be accurately identified so that others associated with the carrier can be immunized. Chronic infection with HBV can be detected by the presence of HBsAg in blood. A major form of HBV transmission is through contaminated blood. Because of resource limitations, many smaller blood banks and transfusion centers in the developing world cannot routinely screen blood for HBsAg with ELISA tests, or they screen with less sensitive and/or specific tests. The high cost of test kits and the dependence on instruments and trained personnel make their widespread use extremely difficult.

Recognition by international health agencies

For many years, the World Health Organization (WHO) and other international health agencies had called for better tools for patient diagnosis, surveillance/monitoring, and blood safety that can be used by community health workers in low-infrastructure settings. In the mid 1980s, USAID anticipated that advances in biotechnology could lead to suitable diagnostic solutions and funded PATH to support research and development. That program, called Diagnostics for Community Health (DiaTech), set the stage for the HealthTech program to embark on an innovative diagnostics development effort starting in 1987. Working groups such as the STD Diagnostics Initiative and Tuberculosis Diagnostics Initiative were organized in the early to mid 1990s as important international health advocates for, and coordination of, the development of needed tools for diagnosis and surveillance of diseases. Currently the Bill & Melinda Gates Foundation supports programs for development of needed TB diagnostic tools through the Foundation for Innovative New Diagnostics (FIND) and support of WHO/Tropical Disease Research (TDR). The Wellcome Foundation as well as the Gates Foundation has also sponsored seminal meetings for discussion of strategies to prioritize, obtain, and accelerate the development of necessary diagnostics. PATH, especially under the HealthTech program, has played a major leadership role throughout these nearly two decades in the implementation of solutions to the problems identified by all these groups.

Technology Solutions/Strategies

Initial discovery/vision

In the early years of HealthTech, PATH embarked on the development of rapid tests for infectious diseases primarily in a solid-phase "dipstick" format. Tests for human immunodeficiency virus (HIV-1 and HIV-2), TB, and hepatitis B were developed and were successfully transferred to developing-country manufacturers. Many are still currently producing and selling these products throughout their regions. The history of dipstick development and introduction is detailed in a separate HealthTech Historical Profile.

In the mid 1990s, PATH realized the dipstick platform was limited in its application. In general it was not optimal for antigen detection assays as it required several steps, which were best performed in a laboratory. Stability was also a concern since the signal reagent needed to be refrigerated for longer shelf life.

IC Strip Test

In 1996, PATH staff focused upon a newer core platform with potential for a wider range of applications—the immunochromatographic strip (ICS) or lateral flow format. This test platform had attractive performance attributes for developing-world application including use of relatively inexpensive off-the-shelf components and reagents, the ability to format the tests for detection of antigens or antibodies, and its usability with a wide range of specimens (e.g., exudates, swabs, urine, serum, plasma, or blood). In addition, PATH observed that once sealed in a pouch, the tests were stable for more than a year, could be shipped without refrigeration, and could be accurately performed and interpreted by minimally trained health care workers. PATH had substantial know-how in developing and optimizing the key colloidal gold signal reagents and their scale-up in volume, based on previous experience in development of dipstick tests

The next six months were devoted to basic platform development using the pregnancy test system, an antigen capture assay for human chorionic gonadotropin (hCG). This was used as a model system since the antibody reagents, specimens, and controls were available and relatively inexpensive, and several commercial test formats could be obtained for comparative evaluation. In this period, materials were identified and signal reagents and buffers were developed. After the hCG test sensitivity and specificity were achieved as equal to or better than the best commercial tests available at that time, the format was judged to be ready to be applied to other applications. This pregnancy test was subsequently transferred to two manufacturers in India.

Design/development of test applications

PATH developed or codeveloped, optimized, and validated a series of simple rapid tests for the target diseases under guidelines that would allow them to be used in conditions typically found in developing countries. For optimal impact, simple and rapid tests were needed that were

economical—or at least cost-effective—for widespread use. The target characteristics of field-appropriate tests include:

- ***Reasonable cost:*** tests that are cost-effective and affordable to public-sector programs, with a reasonable and sustainable profit margin for the producer at an economical scale of production.
- ***Simple:*** minimal training and basic or no equipment needed.
- ***Rapid:*** if used for diagnosis, results available before the patient leaves the clinic—preferably in 10 to 15 minutes or less.
- ***Convenient:*** specimens easy to collect, culturally acceptable, and with minimal preparation or pretreatment.
- ***Stable:*** for potential use in the field or stockpiling at regional centers, the assay should have a long shelf life (one to two years) at ambient temperature.
- ***Accurate:*** appropriately sensitive and specific and able to discriminate past from present (acute) infections.

During this time period a number of commercial companies produced tests for pregnancy and other applications based upon the ICS technology. These tests were characteristically presented in plastic cassettes to increase their perceived quality and keep their tests at a relatively high price. For example, a pregnancy ICS test sold in a cassette could cost US$5 and a chlamydia test US$10. PATH's strategy to achieve affordability included the development of ICS tests in plain strip formats that were operationally and functionally equivalent to tests in cassettes but were much less expensive to make. This approach, in combination with technology transfer to local producers in developing countries enabled the tests to be sold at much lower cost, generally less than US$1.

Syphillis ICS test

PATH's vision was to develop high-priority novel test applications starting with **malaria** and **syphilis** that met optimal requirements for sensitivity and specificity and could be produced and sold by developing-country manufacturers at low cost. Early opportunities also arose to develop tests for **hepatitis B** and **diphtheria**. The ICS tests developed under HealthTech, in general, achieved their design goals. These tests utilized relatively inexpensive, off-the-shelf components such as nitrocellulose membranes, polyester pads, and filter paper, and were formatted to identify specific antigens using monoclonal antibodies, or serum antibodies using specific peptides or recombinant proteins. A typical test could be completed in 15 to 20 minutes and could be performed at the point of care by technicians with minimal training to allow more effective patient follow-up and counseling. Tests for malaria and syphilis were equivalent in accuracy to microscopy or other reference test methods. These tests can be performed directly on whole blood, serum, or plasma specimens taken from patients in rural or smaller clinics or hospitals in the developing world or in resource-limited settings.

After early successes with development and transfer of tests for malaria and syphilis, PATH embarked on the more challenging development of lateral flow tests for gonorrhea and Chlamydia. Development of the gonorrhea test was completed in 2004 and transferred to a manufacturer in India. A Chlamydia test development project was completed in 2005 and is scheduled for transfer.

Validation of the tests by PATH and by third parties

As part of the development process, PATH performed extensive laboratory and field evaluations. Following is a list of the various validation and evaluation activities regarding the PATH ICS tests over the years.

Year	Technology	History and Results
1996	Rapid syphilis test	Informal evaluation with Dr. Subhash Hira and Dr. Stan Eggleston concluded with positive results.
1997	Rapid Chlamydia test	UW samples tested (413); 57% sensitivity; 98% specificity.
1997	Rapid gonorrhea test	DeKalb County samples tested; 83% sensitivity; 85% specificity; additional samples needed.
1997	Rapid gonorrhea test	DeKalb County samples results listed as 92% sensitivity; 91% specificity.
1997	Rapid gonorrhea test	Jefferson County Department of Public Health study: 179 samples; high false positives with endocervical specimens.
1997	Rapid syphilis test	CDC samples tested: sensitivity 98.7%; specificity 88.9%. After resolution of discrepant results by PCR analysis; sensitivity and specificity increased to 100%.
1997	Rapid syphilis test	Initial pilot field evaluation of 200 specimens from STD clinic in Mumbai. “Corrected” data: 100% sensitivity; 96% specificity.
1997	Rapid test for hepatitis B virus	Hepatitis B virus surface antigen assay evaluated on plasma panel supplied by CDC. Specificity 98%-99% compared with ELISA/RIA methods.
1998	Rapid gonorrhea test	British Columbia Center for Disease Control study: 79% specificity (males); 60% from females. Test was reoptimized as a result.
1998	Rapid malaria test	Completed successful field evaluations in Peru and Malawi. These evaluations demonstrated a sensitivity of over 96%, and specificity of 93% or more.
1999	Rapid malaria test	Published malaria ICS data in the World Health Organization (WHO) Bulletin.
1999	Rapid syphilis test	International Society for sexually Transmitted Disease Research (ISSTDR) Conference presentation showed positive results.
2001	Rapid chlamydia test	Data from retrospective study of the CT ICS prototype test using samples from the University of Alabama, Birmingham, was gathered and analyzed. Suboptimal results from this trial indicated need for further test development.

Year	Technology	History and Results
2001	Rapid malaria test	Laos field study data accepted for publication in *Annals of Tropical Medicine and Parasitology*.
2001	Rapid tuberculosis test	Selected sites in Botswana, India, and Ukraine for prospective assessment of test performance.
2002	Rapid gonorrhea test	Initiated and completed a prospective clinical field trial in Johannesburg, South Africa. Demonstrated that the test can achieve acceptable sensitivity and specificity levels with vaginal swabs from women and first void urine from men.
2002-2004	Rapid Chlamydia test	Collected (2002) and evaluated (2004) about 400 urine filtration device (UFD) samples from the South Africa gonorrhea ICS study for retrospective evaluation of the test.
2004	Rapid tuberculosis test	Completed field studies in three sites. Test had low sensitivity but adequate specificity in HIV-positive populations in Botswana. Test had lower sensitivity and specificity in predominately HIV-negative populations in India and Ukraine.
2004	Rapid Chlamydia test	Completed initial analysis of results of the test using clinical samples from Planned Parenthood and demonstrated 60% sensitivity, 98% specificity as compared to strand displacement amplification, a nucleic acid amplification method.
2004	Rapid gonorrhea test	Submitted test prototypes for inclusion in the WHO Sexually Transmitted Diseases Diagnostics Initiative (SDI)-coordinated field trials in Cotonou, Benin. Results were very positive: 70% sensitivity and 98% specificity.
2004	Rapid syphilis test	ICS test tested in a trial by the CDC and showed 100% sensitivity and 96% specificity—an excellent result.
2004	Rapid tuberculosis test	ICS test kits were sent to a CDC-initiated retrospective trial on pediatric samples in Botswana. Results not available yet.
2005	Rapid Chlamydia test	Currently setting up a prospective evaluation of PATH test in Bolivia in collaboration with the Population Council and the Ministry of Health.

Technology Transfer or Licenses

In addition to product development, PATH undertook pilot scale manufacturing of these rapid point-of-care tests. This approach enabled the production of high-quality tests for field evaluation, development of comprehensive master documents for manufacturing and registration of products, complete turn-key technology transfer, efficient training of manufacturing personnel from developing countries, rapid trouble-shooting of post-transfer quality problems, and efficient quality monitoring over the long term.

PATH adopted the policy of licensing production capability of these tests to private-sector manufacturers on a nonexclusive basis. Only diagnostics manufactures supplying developing-world public- or private-sector markets were considered. Licensing agreements with selected recipients addressed the need for diligence in making tests available to public

health programs, accessible to underserved groups, and affordable to resource-poor populations. In order to build capacity for innovation and local product development among recipient manufacturers in developing countries, PATH shared the basic ICS platform and the know-how to develop new test applications. Sometimes the transfers involved codevelopment or collaboration on local adaptation and improvements to the tests.

As just one example of this process, PATH transferred a rapid falciparum malaria test to Orchid Biomedical in Goa, India, together with know-how and training on ICS test development. Subsequently, PATH collaborated with Orchid on the local development of a rapid syphilis test, which was launched in 2001. In 2004, a rapid gonorrhea test developed at PATH was transferred to Orchid. Within a few months, Orchid reported that they had independently improved the sensitivity of the test by thirty-fold and will be sharing those improvements with PATH for subsequent transfer to other countries.

Another strategy is to arrange for several local and international companies to make the tests in order to generate the capacity for large-scale manufacturing, commercial marketing and distribution, and a commitment to making the tests available at an affordable price. For example, PATH developed and validated a syphilis ICS test based on the 17 kD protein, an antigen specific to *T. pallidum*. The test was subsequently licensed to Lee Laboratories (Decatur, GA), a subsidiary of Becton Dickinson (Franklin Lakes, NY), and to J. Mitra & Co. (New Delhi, India). Clinical evaluations of the tests were performed in Peru, South Africa, and Mexico and the data presented at the International Society for STD Researchers conference in 2001. A recent evaluation of the Lee Labs test conducted by CDC showed an excellent result of 100 percent sensitivity and 96 percent specificity.

Following is a list of the recipients of the ICS tests that PATH has initiated and their current status.

Technology	Manufacturer	Country*	Year	Transfer	Sold Units	Still Selling	Current status
Rapid Chlamydia test	Orchid Biomedical	India	2005	Transfer in process	Not yet	N/A	Under negotiation.
Rapid diphtheria test	Orchid Biomedical	India	2001	Transfer	No	N/A	License agreement for antigens not concluded.
Rapid falciparum malaria test	Human GmBH	Germany	2001	Transfer	Yes	Yes	Over 115,000 tests sold.
Rapid falciparum malaria test	Orchid Biomedical	India	1998	Transfer	Yes	Yes	Over 10 million tests sold.
Rapid falciparum malaria test	SPAN Diagnostics	India	1999	Transfer	Yes	Yes	Over 137,000 tests sold.

Technology	Manufacturer	Country*	Year	Transfer	Sold Units	Still Selling	Current status
Rapid gonorrhea test	Orchid Biomedical	India	2004	Transfer	Yes	No	Recently transferred so no sales yet.
Rapid hepatitis B test	Eijkman Institute	Indonesia	1997	Transfer	No	N/A	No sales
Rapid hepatitis B test	J. Mitra & Co. Ltd.	India	1998	Transfer	Yes	Yes	Millions of tests sold/year
Rapid hepatitis B test	Yayasan Hati Sehat	Indonesia	1999	Transfer	Yes	Yes	Not required to report sales
Rapid ICS test technology	Advanced Microdevices (pvt) Ltd	India	1997	Transfer	Yes	Yes	Colloidal gold manufacturing technology for strip tests.
Rapid ICS test technology	J. Mitra & Co. Ltd.	India	1998	Transfer	Yes	Yes	Base technology for IC strip tests.
Rapid ICS test technology	Orchid Biomedical	India	1998	Transfer	Yes	Yes	Colloidal gold manufacturing technology for strip tests.
Rapid ICS test technology	Otsuka Pharmaceutical	Japan	2000	Transfer	Yes	Yes	Base technology for IC strip tests.
Rapid ICS test technology	SPAN Diagnostics	India	1996	Transfer	Yes	Yes	Base technology for IC strip tests.
Rapid ICS test technology	Yayasan Hati Sehat	Indonesia	1999	Transfer	Yes	Yes	Not required to report sales
Rapid pregnancy test	Contech Ltd.	India	1997	Transfer	Yes	No	Discontinued selling due to quality control issues
Rapid pregnancy test	Egyptian Reference Diagnostic Center	Egypt	2000	Transfer	Yes	No	No sales
Rapid syphilis test	J. Mitra & Co. Ltd.	India	2001	Transfer	Yes	No	Quality control issues and access to antigen issues.

Technology	Manufacturer	Country*	Year	Transfer	Sold Units	Still Selling	Current status
Rapid syphilis test	Lee Laboratories	United States	2002	Transfer	No	N/A	Product undergoing extensive US evaluations under CDC supervision. Excellent results.
Rapid syphilis test	Orchid Biomedical	India	2001	Development Collaboration	No	N/A	Technical assistance only.
Rapid syphilis test	Quorum Diagnostics	Canada	1997	Transfer	Yes	No	Went out of business.

* The Program for Introduction of Commercial Technologies for Child and Reproductive Health (PACT-CRH) supported by USAID's India mission facilitated the technology transfer exercises to companies in India.

Policy Environment

The need for new and improved diagnostic tools for priority diseases has been recognized by donors and international agencies and has been supported on several fronts. Advocacy and technology development groups have been formed such as the STD Diagnostics Initiative and Tuberculosis Diagnostics Initiative with secretariats within WHO/TDR. A series of workshops for introduction and quality assurance of rapid malaria tests has been sponsored by WHO/WPRO in Manila and a working group has been formed. PATH was a cofounder of the STD Diagnostics Initiative and has participated in all these initiatives.

International endorsement of rapid tests as best or acceptable practice is an essential step in gaining acceptance and uptake by national health authorities. Appropriate-use policies and guidelines are needed at the international and national (ministry of health) level. This will include advocacy on the use of rapid and simple diagnostics as a national health priority to improve surveillance of disease and individual patient care. It may also involve regulatory changes, promoting education and behavior change among clinicians and health providers, and informing consumers about the uses and advantages of rapid tests. Improved diagnostic practices can be promoted by official endorsement of protocols employing appropriate rapid diagnostic methods to provide a more rapid turn-around time for results to patients and to guide in prescribing drugs. With this objective in mind PATH conducted a workshop in India in the late 1990s sponsored by PACT-CRH, bringing together representatives of industry, state and national governments, regulatory authorities, and nongovernmental organizations (NGOs). The workshop introduced and discussed topics including use guidelines, training, and regulatory issues and policies that would improve current diagnostic practices.

In support of these endorsements and advocacies, it is necessary to build the evidence base for rapid, simple, point-of-care tests as cost-effective interventions in comparison to presumptive or syndromic diagnosis and treatment. Introduction of point-of-care tests will involve new

expenditures by health care programs or shifts in budget from centralized clinical testing facilities to primary patient care facilities. The benefits can accrue directly from the ability of health care workers to diagnose and treat correctly at first contact with the patient. They can also accrue benefits from avoidance of inappropriate use of costly therapies and overuse of antibiotics and other treatments, which result in development of drug-resistant pathogen strains.

PATH carries out cost-effectiveness and market-related studies in order to build the evidence base. For example, two cost studies of the use of rapid syphilis tests in Bolivia and in Mozambique were carried out in collaboration with the Population Council and Health Alliance International respectively and demonstrated the cost/benefits of using rapid tests in comparison with syndromic and laboratory-based diagnosis.

Introduction Phase

Value proposition

A key to effective management of infectious disease is prompt and accurate diagnosis and treatment of patients. In many developing countries, however, resources such as health clinics, trained personnel, diagnostic tools, and therapeutic drugs are often limited at the periphery of the system. As a result, there are unacceptable rates of morbidity and mortality that are associated with use of poor syndromic diagnostic algorithms, lack of diagnostic tools for pathogen identification, poorly focused treatment, and an unjustified or over-use of therapeutic drugs. When accurate, inexpensive, and simple-to-use rapid diagnostic tests are more universally available at the point-of-care they will allow community health care workers to better identify specific infections, make better therapeutic decisions, and immediately deliver treatment. The result is that more patients will receive timely and appropriate treatment; complications are reduced; expensive drugs are conserved; and the overall costs, patient recovery time, and local incidence of infection will all be decreased. From a broader public health perspective, widely available point-of-care tests will reduce the inappropriate use of medications that are causing the emergence of drug-resistant pathogens.

Mainstreaming/general acceptance

Of the several tests developed at PATH under the HealthTech program, the malaria test has achieved the widest uptake to date. Rapid point-of-care malaria tests are becoming well established as useful and valuable tools for diagnosis of patients and for other uses in the field. Following the introduction of the PATH-developed test, several commercial manufacturers have developed high-quality falciparum malaria test kits resulting in very competitive prices. Many are based on PATH's original design, focused on falciparum malaria only, and marketed as plain strips rather than cassettes, and are therefore comparatively inexpensive. A few pan-malaria tests are now available and able to detect all four types of malaria. These are useful and recommended in regions where there are multiple species present, though they tend to be expensive compared to falciparum tests.

The falciparum-only malaria tests are finding wider acceptance beyond continental Africa, where falciparum malaria predominates. They are also being used effectively in regions where there are mixed types of infections, since there are potential cost savings if a patient does not have

falciparum malaria. In such resource-limited situations, a positive falciparum malaria test from a patient with fever would indicate that a first-line therapeutic such as artemisinin is administered, whereas a patient that tested negative may be given chloroquine, a much less expensive, yet effective, drug for non-falciparum malaria. WHO is advocating and using rapid malaria tests in several large programs it sponsors, but one concern at present is monitoring their quality and dependability in the field.

Other rapid tests such as those for syphilis have been accepted for use in some situations, such as in antenatal clinics, but not in all situations since they detect both past treated and current infections. They are therefore less useful in populations where a high prevalence of disease is expected, such as in patients attending STI clinics, but are very useful in, for example, screening pregnant women from the general population.

ICS quality control

Other rapid tests such as those for gonorrhea are relatively new and as a result are still in the early adopter stage of uptake. Few companies have attempted to develop these tests since the incidence of gonococcal infection has been declining in many developed countries. There have also been considerable technical difficulties in designing a test, the need to use a relatively invasive sample and the high sensitivity and specificity to compete with nucleic acid amplification methods. The recent positive results for the PATH-developed GC test in a WHO evaluation are exciting in that few companies have achieved this level of sensitivity and specificity. In addition, PATH's transfer recipient reports that they have improved the sensitivity thirty-fold since they took over production responsibility. Soon the test will be on the market as one of the few, rapid, inexpensive tests, for diagnosis of gonorrhea in women with symptomatic and asymptomatic infections in developing countries.

Commercial rapid Chlamydia tests have been available now for several years, but they have been expensive and their overall sensitivity is relatively low compared to other test methods such as nucleic acid amplification, now used by many reference laboratories. However, studies have indicated that in the developing world a significant proportion of patients are lost to follow-up if they are asked to return to the clinic on another day to obtain their test results and receive treatment. Some reports have indicated that a rapid test providing same-day results but at a lower sensitivity would actually provide greater benefit given that more patients with disease would be treated rather than lost to follow-up. The PATH-developed Chlamydia test targets this application but also aims to improve sensitivity and be much more affordable compared with

currently available commercial tests. Transfer of this test to an Indian producer is currently underway.

Hurdles/Constraints/Limitations

Rapid tests are simpler to perform and interpret than laboratory-based reference method assays since they do not require electricity, special equipment, or extensive training. Health workers can quickly learn to perform the tests and require infrequent retraining. PATH has succeeded in technically adapting the ICS platform for various diseases. However, some rapid tests are still not sufficiently sensitive or specific for accurate use at the point of care. For example, PATH's malaria tests only detect *P. falciparum* and will therefore lack sensitivity in regions where other species of *Plasmodium* are found. They also cannot be used effectively in hyperendemic areas where the majority of individuals may have low titers of circulating *P. falciparum* antigens. Antigens may persist in circulation even after successful therapy and could produce confusing results with *P. falciparum* tests when assessing treatment failure or drug resistance.

Operationally, when rapid tests are used in the clinic, the role of a central laboratory is diminished and there is a real or perceived loss of control over the data. As rapid test kits are accepted for clinical use, submissions of specimens to central health authorities for viral isolation and typing have been predicted to decrease. Since the available rapid tests do not distinguish novel virus subtypes from known subtypes, fewer viral isolates can be obtained and, as a result, the capacity for early detection of new variants may be reduced.

Operational difficulties with rapid point-of-care tests may include:

- Integrating the tests into routine clinical procedures, since the time each patient receives with a provider is often very limited in already overburdened rural clinics.
- Purchase and use of rapid tests may not be possible if the ministry of health (MOH) does not provide a budget, cannot afford them, or does not believe they are cost-effective.
- Establishing a quality assessment system to ensure the accuracy of rapid tests, since there is always the possibility that the tests, even though they are robust, may degrade if subjected to extreme environmental conditions in transport or storage before use.
- More accurate diagnostic testing will result in fewer patients getting drugs for treatment, in contrast to syndromic protocols now in use. Patient confidence in their providers may erode if they do not receive treatment when it is a custom to expect something.

There are other public perceptions of rapid test technologies that need to be corrected or overcome. For example, PATH personnel have noted that patients and/or their health care providers may not believe the results of a rapid test because it seems to be too fast and too easy to perform. Patients also may not trust local health workers to perform testing at local clinics because the results may not be perceived as accurate compared with tests performed in remote laboratories

These are all examples of the challenges of introduction of new technologies that disrupt previously established ways of doing things. It will require the attention and effort of many

organizations over many years to establish point-of-care diagnostic testing as a standard practice for primary health care in the developing world. PATH's role in point-of-care diagnostics continues to be the development of tests that address priority needs in developing-country health care and that set the standard for affordability and ease of use. PATH has also taken the opportunity to build developing-country capacity for innovation in diagnostics because local manufacturers will be a principal source of promotion and supply as interest and demand for these products grows. The lateral flow immunochromatographic platform is hard to beat as a robust, simple, and low-cost self-contained means to detect a wide variety of diseases. PATH continues to seek funding and collaboration to overcome some of the limitations in sensitivity or specificity of lateral flow tests for certain important diseases like tuberculosis. Several new enhancements are showing promise in this regard and are likely to extend the utility of this format well into the future, even as the new "lab on a card" platforms emerge and find their place in developing-world health care. PATH also feels that it is important and urgent to continue to build the evidence base for cost-effectiveness and public health benefit of rapid, point-of-care tests and is seeking support from funders and collaborators to accelerate this "mainstreaming" process.

Evidence of Impact

As with most innovations in developing-world public health care programs, the uptake of rapid point-of-care tests is slow, occurring over years and often extending into one or two decades. The concentrated resources and focused effort characteristic of product introduction in the commercial sectors of the industrial world are generally absent from the process. Earlier impact has to be measured in terms of progressive indicators—uptake by producers, early adopters, influential endorsements, independent field evaluations, number of tests sold, spread of product to new countries, and emerging competitive tests. Based upon these early indicators, there is no doubt that HealthTech's efforts in developing simple and rapid tests have had a considerable impact on the efficiency and cost of diagnosis for selected infectious diseases and have saved many lives and/or prevented progression of disease. For example, there are currently several million rapid tests for falciparum malaria now being used globally as the direct and indirect results of PATH's efforts. However, it will be a few years before their true impact on the control of malaria and their relative programmatic contribution to reduce the impact of disease can be fully known. In the interim, there are a few indirect indicators of impact, including the following.

Sales Data

One indicator of the potential widespread use of the tests that PATH has developed is to track the sales of the products by the manufacturers who are producing and selling them. Recent reports from those companies for some but not all tests are presented in the following table, indicating that millions of tests are reaching the target populations.

PATH Test	Year Transferred	Years Sold	Manufacturer	Units Sold to Date	Distribution Area
Rapid gonorrhea test	2005	2005	Orchid	0	No sales yet.
Rapid hepatitis B test	1998	1999–2004	J. Mitra	Millions	worldwide
Rapid hepatitis B test	1998	1998–2001	Orchid	3,355,965	worldwide
Rapid hepatitis B test	1999	1998–2003	Yayasan Hati Sehat	530,070	worldwide
Rapid malaria test	1998	1998–2002	Orchid	10,196,890	worldwide
Rapid malaria test	1999	2000–2004	SPAN Diagnostics	137,289	worldwide
Rapid malaria test	2001	2002–2004	Human	115,000	worldwide
Rapid pregnancy test	1997	1999–2004	Advanced Micro Devices	3,112	worldwide
Rapid pregnancy test	1998	1998–2001	Orchid	31,091,098	worldwide
Rapid pregnancy test	1999	n/a	Otsuka	n/a	worldwide

Stimulation of Competition

The following anecdotes describe the stimulation of competition through the availability of rapid tests produced by PATH and its collaborators:

- PATH, through HealthTech and the PACT-CRH program in India, has worked with key members of the Indian diagnostics industry to provide them with quality applications, procurement of key antibodies and reagents, know-how for assay development, and especially in efficient volume-production of colloidal gold signal reagent. These advantages have allowed the Indian manufacturers to produce higher-quality products at low prices that are able to meet the needs of local and export health care markets in the public and private sector.
- PATH's policy has been to develop and transfer ICS test platform technology and its applications on an unrestricted, nonexclusive basis to diagnostics manufacturers and provide technical assistance to others that are already versed in the art. For example, the

falciparum malaria test was transferred to manufacturers in India and Germany, and PATH also provided technical assistance to other companies in India and South Africa. With several companies all producing similar products, competition has been encouraged, resulting in affordable pricing in both the public and private sectors.

- PATH has also worked with international health agencies in advancing and introducing the entire range of rapid malaria tests available from commercial competitors into use in developing countries.
- A website set up by PATH (http://www.rapid-diagnostics.org) lists current commercial manufacturers and distributors of selected rapid tests such as hepatitis B, malaria, and syphilis, as well as provides links and evaluation results, thereby giving readers information on the range of tests that are commercially available and how to access them.

Publications

Another method of assessing the widespread awareness and knowledge of PATH's work in this area is through a scan of the publications that have been published on these topics. Since PATH started work on the ICS test platform in 1995, the following articles have been published in peer-reviewed literature and presentations at professional and academic meetings.

Year	Technology	History and Results
1998	Rapid malaria test	Loutfy MR, Assmar M, Hay Burgess DC, Kain KC. Effects of viral hemorrhagic fever inactivation methods on the performance of rapid diagnostic tests for *Plasmodium falciparum*. *Journal of Infectious Disease.* 1998;178(6):1852–1855.
1999	Rapid diagnostic tests	Tam MR. Laboratory diagnosis of sexually transmitted diseases in resource-limited settings. Chapter 103, pp 1409–1420. In *Sexually Transmitted Diseases* Third Edition. McGraw-Hill publishers. Copyright © 1999. Holmes KK, Mardh PA, Sparling PF, Lemon SM, Stamm WE, Piot P, Wasserheit JN, Editors.
1999	Rapid malaria test	Mills CD, Hay Burgess DC, Taylor HJ, Kain KC. Evaluation of a rapid and inexpensive dipstick assay for the diagnosis of *Plasmodium falciparum* malaria. *Bulletin of the World Health Organization.* 1999; 77(7):553–559.
2001	Rapid malaria test	Labbe AC, Pillai DR, Hongvangthong B, Vanisaveth V, Pomphida S, Inkathone S, Hay Burgess DC, Kain KC. The performance and utility of rapid diagnostic assays for *Plasmodium falciparum* malaria in a field setting in the Lao People's Democratic Republic. *Annals of Tropical Medicine and Parasitology*. 2001;95(7):671–677.
2002	Rapid diphtheria test	Engler KH, Efstratiou A, Norn D, Kozlov RS, Selga I, Glushkevich TG, Tam M, Melnikov VG, Mazurova IK, Kim, VE, Tseneva GY, Titov LP, George RC. Immunochromato-graphic strip test for rapid detection of diphtheria toxin: description and multicenter evaluation in areas of low and high prevalence of diphtheria. *Journal of Clinical Microbiology.* 2002;40(1):80–83.

Year	Technology	History and Results
2002	Rapid chancroid test	Patterson K, Olsen B, Thomas C, Norn D, Tam M, Elkins C. Development of a rapid immunodiagnostic test for *Haemophilus ducreyi*. *Journal of Clinical Microbiology.* 2002;40(10):3694–3702.
2002	Rapid syphilis test	Zarakolu P, Buchanan I, Tam M, Smith K, Hook III EW. Preliminary evaluation of an immunochromatographic strip test for specific *Treponema pallidum* antibodies. *Journal of Clinical Medicine.* 2002;40(8):3064–3065.
2003	Rapid gonorrhea test	The data generated through the South African field trial were presented at the ISSTDR, July 2003.
2004	Rapid tuberculosis test	Talbot EA, Burgess DCH, Hone NM, Iademarco MF, Mwasekaga MJ, Moffat HJ, Moeti TL, Mwansa RA, Letsatsi P, Gokhale NT, Kenyon TA, Wells CD. Tuberculosis serodiagnosis in a predominantly HIV–infected population of hospitalized patients with cough, Botswana, 2002. *Clinical Infectious Diseases*. 2004;39(1):1–7.
2005	Rapid gonorrhea test	The data generated through the Benin field trial will be presented at the ISSTDR, July 2005.

Third Party Comments

The following are references to PATH tests in various articles.

Technology	Reference	Citation
HealthTech	PATH's Technologies for Health (HealthTech) Project has the goal to improve child survival and maternal health through practical, affordable, appropriate technologies. PATH collaborates with organisations to develop, advance or adapt technologies that address PHC needs in resource-poor countries.	**The Public Health Care Laboratory.** The website for laboratory personnel in developing countries. http://www.phclab.com/Addresses/PATHad.htm (Accessed June 8, 2005).
Rapid diagnostics website	"Strives to provide information useful to international public health professionals. Country-level program managers, medical providers, and policy decision-makers can visit this informative site to educate themselves about rapid diagnostic technologies appropriate for developing countries and other resource-limited settings. This website is managed by the Program for Appropriate Technology in Health (PATH) and its development supported by USAID under the HealthTech IV program."	**From the Global Health Council** http://www.globalhealth.org/news/article/2370. (Accessed June 8, 2005).

Technology	Reference	Citation
Rapid diagnostics website	PATH site aiming to provide information on RDTs for tropical diseases to public health program managers and decision-makers in developing countries.	**WHO/WPRO** http://www.wpro.who.int/rdt/link9.asp (Accessed June 8, 2005).
HBsAg IC Strip Test	This rapid test technology was developed at PATH and then transferred to a manufacturer in India. In this study, it was concluded to be of use in laboratories without adequate infrastructure for gold standard serological tests.	Raj AA, Subramaniam T, Raghuraman S, Abraham P. Evaluation of an indigenously manufactured rapid immunochromatographic test for detection of HBsAg. *Indian Journal of Pathology & Microbiology.* 2001;44(4):413–414.
HBsAg IC Strip Test	A two-year prospective study in India evaluated this test as compared to the gold standard and found the sensitivity and specificity to be adequate.	Kaur H, Dhanao J, Oberoi A. Evaluation of rapid kits for detection of HIV, HBsAg and HCV infections. *Indian Journal of Medical Sciences.* 2000;54(10):432–434.
Malaria IC Strip Test	This study evaluated molecular amplification, microscopy, and the PATH-developed rapid test for malaria diagnosis. In areas where microscopy expertise is lacking and laboratory capacity limited, the rapid test is considered useful.	Parkes R, Lo T, Wong Q, Isaac-Renton JL, Byrne SK. Comparison of a nested polymerase chain reaction–restriction fragment length polymorphism method, the PATH antigen detection method, and microscopy for the detection and identification of malaria parasites. *Canadian Journal of Microbiology.* 2001;47(10):903–907.
Gonorrhea and Chlamydia IC Strip Tests	The development of rapid diagnostic tests for gonorrhea and Chlamydia infections by PATH, with support from USAID, UNFPA, and the Bill & Melinda Gates Foundation, can prevent infertility by functioning as screening tools in rural clinics and small hospitals.	Cates W. Preserving fertility: an underappreciated aspect of sexual health. *FHI Network.* 2003;23(2).
HealthTech, Uniject, and Malaria IC Strip Test	In this electronic newsletter from the World Bank, the Technologies for Child Health (HealthTech) project, managed by PATH and funded by USAID is praised by the First Lady. Uniject field trials in Bolivia and Indonesia are also presented as is the rapid, simple Malaria IC Strip Test.	What's New in the PHNFLASH Archive? Electronic Newsletter on Population, Health and Nutrition Issues. Population, Health and Nutrition (PHN) Department, World Bank. May 1995.

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