

Measurement of Biomarkers in Surveys in Developing Countries: Opportunities and Problems

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RELIABLE AND COMPREHENSIVE data on disease levels, patterns, and trends in populations are required to monitor global and local epidemics and to assess the effectiveness of public health approaches to the prevention and control of disease and injury. For most developing countries, little is reliably known about causes of mortality or about disease incidence and duration. For these populations "epidemiological manipulation, extrapolation, and estimation procedures" have been used to make imprecise estimates of the burden of disease (Murray and Lopez 1996), and meager databases are used to plan and evaluate health programs. Advances in technology offer the opportunity to include biological and clinical data collection in existing large-scale, national sample surveys. Such data could result in significantly better insight into public health problems and more rational and equitable policies leading to improved health in developing countries. Collecting biological and clinical data—referred to here as biomarkers—in large-scale national surveys, however, poses a number of logistical and ethical issues that must be successfully addressed.

Existing public health measurement

The assessment of health status in populations of most developing countries is based on the limited data available from epidemiological research,

health facility reports, and national surveys. Epidemiological research, an important data source, is often focused on a single disease and conducted in relatively small, selected populations. Health facility statistics generally have a broader coverage and may include disease surveillance, morbidity reports, and service provision data (e.g., vaccination). In many countries, however, the usefulness of such statistics is limited because of low health-services use for many diseases and conditions and because such data are often incomplete, untimely, and insufficiently accurate to describe the burden and distribution of disease in populations.

National population-based health interview surveys provide data on mortality levels, differentials, and trends, causes of death (through "verbal autopsy"), and self-reported morbidity. Examples include the USAID-sponsored Demographic and Health Surveys program (DHS), UNICEF's Multiple Indicator Cluster Sample surveys (MICS), and the Reproductive Health Surveys of the Centers for Disease Control and Prevention (CDC). These surveys have traditionally focused on reproductive and child health, although adult men increasingly are included in the DHS and CDC surveys. Biological and clinical data collection in these surveys has generally been limited to anthropometry. Recently, however, some of the Demographic and Health Surveys included assessment for anemia of women of reproductive age and young children (Sharmanov 2000). This is done by means of drawing a small amount of capillary blood and the results, read by a portable, battery-operated digital device, are available for in-the-field recording and feedback to the survey participants. Several recent MICS-related surveys included dried blood spots to assess tetanus antitoxin and retinol (vitamin A) levels. Health examination surveys have been conducted, for example, in Pakistan and Egypt and included a wide range of biological and clinical data collected at a mobile or stationary clinic in a survey cluster (Fisher, Pappas, and Limb 1996). These surveys tend to be costly because of the need for extensive involvement of medical personnel and the duration of the investigation (2–3 years). Many developing countries conduct other types of national surveys, which may include health data collection.

Even if health information systems based on routine data from health facilities in developing countries are significantly improved, the need will continue for national population-based surveys that provide data on health-related attitudes, behaviors, and self-reported health status. In developed countries with extensive routine health information, regular population-based surveys with health interviews and biomarker data collection are considered necessary. For instance, the National Health and Nutrition Examination Survey (NHANES) in the United States examines a nationally representative sample of about 5,000 persons each year and includes biomarkers for numerous diseases and conditions. Incorporating biomarkers in national surveys will provide relevant disease prevalence data, which are nationally or subnationally representative, by measuring and tracking the

burden of disease (including subclinical disease) rather than self-reported illness. It will also allow assessment of the relationship between disease on one hand and risk factors, treatment interventions, and prevention programs on the other.

Technological advances

Recent technological advances offer rapid, relatively inexpensive, and logistically feasible diagnostic tests that could be incorporated into large-scale, population-based surveys conducted even in remote areas of developing countries. Paramedical or nonmedical personnel can collect certain types of body fluids in field conditions. Thus, for example, disposable lancets with a calibrated self-retracting incision device make it feasible to obtain capillary blood from the fingertip or heel during household surveys. An absorbent pad can be placed between the lower gum and cheek for two minutes to collect oral mucosal transudate. Self-administered vaginal swabs have been acceptable collection devices in some research studies in developing countries (Wawer et al. 1999). The use of dried blood spots on filter paper and the use of saliva samples circumvents a major logistical stumbling block in developing countries—the need for a “cold chain” from field to laboratory. In addition, these techniques greatly lower the risk of exposure to infectious agents for the survey team, study participants, and community members.

The numbers of assays that can be performed on a dried blood spot or oral fluid sample have greatly increased in the last few years. More and better tests are likely to become available in the near future. Assays based on oral specimens can be used for the diagnosis of a range of infectious diseases (antibody-based diagnosis—e.g., HIV1 or HIV2, hepatitis A, B, or C, *Helicobacter pylori*, measles, mumps, rubella, syphilis, and cytomegalovirus) and some chronic diseases (autoimmune disorders such as Sjögren's syndrome, rheumatoid arthritis, myasthenia gravis, diabetes type I and II) (George and Fitchen 1997). Additional tests can be performed on dried blood spots, including all tests that can be done on oral fluid and several biomarkers of micronutrient deficiencies (serum retinol, hemoglobin, folate, zinc metallothionin, thyroid stimulating hormone), and some tropical diseases (malaria). Some of the tests can be done in the survey participant's home (e.g., hemoglobin); others require a laboratory. Urine samples can be used to test for several sexually transmitted infections, using polymerase or ligase chain reaction (PCR and LCR), although both tests require a well-functioning cold chain from field to laboratory.

It is possible that the addition of specimen collection will increase rates of nonresponse in surveys. Research studies and DHS surveys that have collected capillary blood for anemia testing have so far not recorded any major increase in nonresponse bias. Feedback from data collection teams

has indicated that most respondents were actually keen on having the test. Collection of less-invasive specimens such as saliva or urine may help minimize nonresponse, along with providing test results and appropriate counseling. Differential changes in nonresponse (e.g., more men refusing participation than women) need to be evaluated carefully to assess and possibly adjust for biases.

Table 1 summarizes issues in the measurement of disease incidence and prevalence through survey questions and biomarkers in selected areas of health. In most areas health interviews are not able to provide accurate data on the prevalence or incidence of specific diseases, although they provide background and risk factor information. The degree to which biomarkers can contribute to better measurement depends on diagnostic test characteristics (sensitivity, specificity, costs) and the extent to which it is possible to obtain an accurate sample. Some tests require a cold chain from field to laboratory testing, which greatly complicates survey execution and increases the risk of inadequate or inaccurate samples for testing. Also, many tests cannot distinguish between past and recent exposure to infection (e.g., HIV, HSV-2, measles) so that incidence cannot be measured. A recent innovation in the field of HIV is the use of two assays with different sensitivity from the same specimen. A positive result from the sensitive test but a negative result from the less-sensitive test is indicative of early infection in adults (Janssen et al. 1998). This sensitive/less-sensitive testing strategy can be used to estimate incidence in surveys, but sample size will have to be large and general population incidence fairly high.

Justification for measurement of biomarkers

The inclusion of a biomarker in population-based surveys can be considered if the biological condition is expected to be sufficiently common in the general population (e.g., if prevalence is likely to exceed 5 percent) and if the condition is considered to be of public health significance. In that case biomarker information can be used to assess levels and distribution of the condition in the population, to improve understanding of the mechanisms affecting ill health, and potentially to improve health programs.

The decision on whether to include biological and clinical data collection in surveys rests on more than feasibility and cost and varies by health area. To add value to existing data, the inclusion of biomarkers in national surveys must improve on biological data already available from other sources. Data from biological specimens must also improve on data that might be obtained from a survey without the collection of specimens, for example through clinical diagnosis or self-reporting of symptoms. Finally, it would be inappropriate to collect such data if the resulting information is unlikely to influence policy decisions.

The extent to which the inclusion of biomarkers in surveys adds to existing data depends largely on the availability and representativeness of existing data. Population-based surveys have many advantages over clinic population data for assessing the size and distribution of a problem and for subsequent planning and evaluation of health interventions. Assessments of micronutrient deficiencies and HIV prevalence provide illustrative examples. The prevalence of mild and moderate levels of micronutrient deficiencies, such as vitamin A deficiency and anemia, can be measured accurately only through biological data collection. Research during the past decade has shown that mild to moderate deficiency may lead to increased risks of morbidity and mortality, and knowing population prevalence and distribution may have benefits for health planning and policy. In most countries with severe AIDS epidemics, national trends in HIV prevalence are monitored through antenatal clinic-based surveillance. There are, however, numerous limitations related to such surveillance. In most countries, few surveillance sites are found in the smaller antenatal clinics in the rural areas where often the majority of the population lives. Multiple biases may affect levels and trends in HIV prevalence observed in antenatal clinics because of low attendance by pregnant women, selection bias for sexual activity, and the fertility-reducing effect of HIV infection. This source of surveillance provides limited data on the distribution of the disease by various population categories, such as socioeconomic status, and no data on men. We have little experience to demonstrate how much gain can be achieved by measuring HIV prevalence in population-based surveys. Potentially, however, HIV prevention and AIDS care programs could benefit substantially from having more accurate data on levels, trends, and differentials in HIV prevalence.

The contribution of biomarker measurement in surveys also depends on existing or potential health programs or policies. The decision to provide women with postpartum vitamin A supplementation may be made on the basis of serum retinol levels from dried blood spots collected during a population-based household survey. Measuring antitoxin levels, based on dried blood spots, in women who recently delivered may accurately assess tetanus vaccination coverage and help evaluate the quality of the vaccines and the immunization program. Population-based data on the prevalence of antibodies to vaccine-preventable diseases may lead to the (re-)introduction of vaccines or changes in immunization policies with regard to booster doses or age at vaccination. Biomarker data on sexually transmitted diseases may indicate high prevalence of symptomatic and asymptomatic infections. Only population-based surveys using biomarkers can provide adequate data on the actual spread of various pathogens in populations; these data could be used to design and monitor prevention and treatment programs. For instance, health interview surveys have not been successful in measuring

TABLE 1 Measurement of the burden of disease through survey questions and biomarkers in selected areas of health

Disease/health area	Measurement through interview	Biomarker/diagnostic test	Sample needs	Comments	Selected references
HIV infection	Not feasible if asymptomatic; even with symptoms difficult	HIV antibody test, positive 4–6 weeks after infection; rapid tests on the spot or in lab; HIV antigen detection through nucleic acid amplification techniques (e.g., PCR, LCR)	Serum; blood-spotted filter paper; saliva; urine	Many tests available for antibodies, extensively used in surveys; HIV detection through DNA amplification tests requires cold chain to transport sample and special lab facilities	Gallo et al. 1997 Giles et al. 1999 Fylkesnes et al. 1998 Grosskurth et al. 1995
Syphilis	Presence of genital ulcer in last 12 months or at the time of interview—low validity of responses	Non-specific test (e.g., RPR) for recent or current infection; specific test (e.g., TPHA, FTA-Abs) for past or recent exposure	Serum; saliva-based test under development	Often a combination of a non-specific and specific test is used to make the diagnosis of syphilis; RPR or Trust can be done in the field. Specific tests need lab; RPR test used in antenatal clinics	Wawer et al. 1999 Mayaud et al. 1997
Gonorrhea/chlamydia trachomatis infection	Presence of genital discharge in last 12 months or at the time of interview—low validity of responses	LE urine dipstick; Nucleic acid amplification tests (PCR, LCR)	Urine	Dipstick has low sensitivity and specificity; PCR/LCR detect current infections, but require cold chain and special lab facilities	Tyndall et al. 1999 Wawer et al. 1999
Herpes simplex virus infection (HSV-2)	Presence of genital ulcers in last 12 months or now—low validity of responses	HSV-2 antibody test	Serum; blood-spotted filter paper	As with all antibody tests, gives information about ever being exposed to HSV-2; HSV-1 antibody test also available	Obasi et al. 1999
Trichomoniasis	Presence of genital discharge in last 12 months or now—low validity of responses	Culture of trichomonas vaginalis; PCR in urine	Self-administered vaginal swab; urine	Swabs give good results, and high participant acceptability has been reported; PCR is much more costly, requires cold chain and special lab facilities	Wawer et al. 1999

Vaccine-preventable diseases	History of disease; home-based vaccination record (health cards) or recall of vaccinations by main caretaker	Serological tests for antibodies against rubella, measles, mumps, diphtheria, and tetanus antitoxins	Serum; blood-spotted filter paper; saliva	Cannot distinguish infection-induced antibodies from vaccine-induced antibodies	Perry et al. 1993
Malaria	Presence or recent history of fever; poor specificity	Rapid diagnostic test, dipstick format to detect parasite	Whole blood; blood-spotted filter paper	Most tests perform very well for high parasite density infections; less sensitive and specific for low parasite density infections; quick results	Makler et al. 1998 Mills et al. 1999
Anemia	Presence of pallor and other anemia symptoms; poor validity	Portable Hemocue test; WHO color scale	Capillary blood obtained by finger prick (Hemocue); blood-spotted filter paper (color scale)	On-the-spot results; now used in many national surveys	Sharmanov et al. 1998
Vitamin A deficiency	Night blindness or eye symptoms	Test for serum retinol	Serum; blood-spotted filter paper	Test needs to be done in lab, gives fairly good estimates at population level, but less so at individual level; field test result reader under development	Tanumihardjo et al. 1996
Iodine deficiency disorders	Presence of goiter, mental retardation	Salt testing for iodine content; urinary iodine excretion	Household salt; urine	Salt testing extensively used in household surveys (e.g., UNICEF surveys)	May et al. 1997
Cardiovascular diseases	Medical history, anti-cardio-vascular drug use	Blood pressure; anthropometry; multiple markers of cardiovascular health, musculoskeletal health, respiratory function, metabolic processes etc.	Portable blood pressure and anthropometric equipment; serum	Used in adult health examination surveys	Crimmins and Seeman 2001; Fisher et al. 1996

FTA-Abs = fluorescent treponemal antibody absorption test

HSV = herpes simplex virus

LCR = ligase chain reaction

LE = leukocyte esterase

PCR = polymerase chain reaction

RPR = rapid plasma reagin

TPHA = *Treponema pallidum* hemagglutination assay

morbidity attributable to malaria. The development of rapid diagnostic malaria tests has great potential to better measure the occurrence of this disease and to improve and evaluate interventions. In many populations in developing countries, the epidemiological transition has progressed and non-communicable diseases have become much more prominent. Biomarker data collection in surveys provides an opportunity to assess the prevalence of these diseases, including hypertension, obesity, and cardiovascular disorders, and the occurrence of disabilities. These data can be used to design appropriate preventive and therapeutic interventions. On the other hand, biomarkers are not available for some conditions. For example, assessment of the prevalence of mental illness would have to rely on a series of standardized questions (Derogatis and Melisaratos 1983).

Logistical challenges

The development of rapid diagnostic tests that use saliva, urine, vaginal secretions, or dried blood spots collected on filter paper reduces many logistical constraints and safety concerns associated with traditional tests and specimen collection methods. Nevertheless, some safety, logistic, and quality issues remain to be addressed. The risk of pathogen transmission must be minimized through proper handling and disposal of specimens and waste. This requirement implies proper training of survey teams and laboratory personnel, as well as use of appropriate methods for specimen collection and waste disposal.

While supplies and equipment to collect specimens, such as dried blood spots or saliva, may be inexpensive and easily procured for the survey, laboratories in developing countries may not be equipped to conduct some of the assays. Tests that require specimens to be shipped to laboratories outside the country may increase costs and encounter political obstacles. Efforts should be made to promote the transfer of technology to developing countries so that the tests can be run in national or regional laboratories and to develop tests that can be carried out in the field.

Regardless of where the assays are conducted and no matter how simple to perform, diagnostic tests require quality control related to storage of supplies and equipment, specimen collection, handling, and storage, as well as to conducting the test itself. Retesting of samples in an optimal setting in a different laboratory should be part of the procedure. Failure to maintain quality controls throughout may produce spurious results that lead to incorrect conclusions and inappropriate policy and program decisions. Geographically dispersed surveys with multiple survey teams require careful training and regular supervision to ensure standardization of procedures and maintenance of equipment in difficult situations, such as areas with shortage of electricity and voltage spikes, high ambient temperature, dust, and few spare parts or repair facilities.

Ethical challenges

Even for health interview surveys that do not include measurement of biomarkers, procedures should already be in place to secure approval by host- and donor-country ethical review boards, obtain informed consent from participants, and ensure confidentiality. Before protocols can be submitted to review boards and consent statements can be written, decisions must be made regarding the storage of specimens for future testing. These decisions include: which organization has responsibility for storage; what are the criteria to determine when, by whom, and for what reasons future testing will be allowed; who will have access to the results; and whether the stored specimens are unlinked and anonymous or linkable to the survey data. If specimens are linkable to identifiers, decisions must be made regarding obtaining informed consent and providing counseling or treating participants whose specimens will be tested in the future. Linkable data could potentially be misused to discriminate against individuals on the basis of their health or genetic profile. Deciding against specimen storage obviates the need to make these decisions.

The greatest challenge presented by adding biomarkers is what to do about advising people of their test results and treating them when necessary. Traditionally, survey teams have not provided or recommended treatment or preventive action to respondents who report illness or high-risk behavior. Even when more objective data, such as anthropometric measures or vaccination status based on health cards, identify malnutrition or inadequate vaccination coverage, mothers have not been referred to the health care system or even told that their child is malnourished or inadequately vaccinated.

In sharp contrast to local research studies, the nature of large-scale sample surveys makes it virtually impossible to provide feedback to the respondents if no accurate, rapid test can be done on the spot. Only if an excellent communication network exists for all individuals could survey participants, in principle, receive feedback on the test results, if they choose to be informed. Few developing countries have such an infrastructure that also ensures confidentiality. If a rapid test can be carried out in the field, there are two options. The individual can be given feedback and referred to a nearby health facility for treatment or advice, or counseling and treatment can be given on the spot. For example, options for counseling and treating anemic survey respondents include: recommendation of appropriate foods; provision of a one-month supply of iron with or without multivitamins; referral to the local health facility; and obtaining permission to notify the local health workers of the results.

Any decision to provide treatment within the context of a survey would add to training, transport, and commodity costs. It also raises questions about accuracy of diagnosis. Some tests provide reliable results at the population

level but less reliable results at the individual level; others require confirmatory tests. For some conditions this requirement is not a drawback since an incorrect diagnosis of or even unnecessary treatment for anemia or vitamin A deficiency is unlikely to have negative consequences. An incorrect result for other conditions, such as sexually transmitted infections or HIV, is of far greater concern. The need for confirmatory laboratory testing makes it difficult to follow up individuals for counseling or care, and the potential for breaches of confidentiality is high. Therefore, referral to a nearby health facility for treatment or advice appears to be the most viable option, assuming that appropriate services are available at the facility.

Special consideration needs to be given to including HIV testing in population-based surveys. AIDS is as yet an incurable disease that is highly stigmatized in most societies. A positive test result can have severe consequences for an individual. One option for surveys is a variation on unlinked, anonymous HIV testing used in HIV surveillance in health care settings. Blood or other specimens are obtained, with the consent of the respondent, for non-HIV testing (e.g., syphilis or anemia testing). Leftover samples are tested for HIV in laboratories after delinking the sample from all identifying questionnaire information (such as name, identification number, and cluster of the sample). Only a few key characteristics (such as general geographic area of residence, sex, age, parity, level of education) are retained with the sample, so that tracing back the test result to the individual becomes impossible.

Conclusion

There is little doubt that technological advances have opened the door for better assessment of health status and the burden of disease in human populations. National surveys—regularly carried out in many countries—are an important vehicle for the application of these technological advances, especially in countries with limited and defective data on health and disease. Large-scale population-based surveys are of particular importance in the assessment of inequalities in health and health status. Measurement of biomarkers per se in surveys could result in significantly better insight into public health problems and more rational and equitable policies leading to improved health. The combination of traditionally collected behavioral data with biological and clinical data affords many possibilities to better assess health problems and to develop the most cost-effective set of interventions. Careful assessment and discussion of the potential public health benefits, ethical issues, and logistical challenges should guide the application of technological advances in population-based surveys.

Note

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