DATA DOCUMENTATION
LABS

Specimen Collection

Blood and urine specimens were collected at the clinic from consenting survey participants. All specimens were processed according to protocol, and aliquots were shipped to various laboratories to be analyzed. The blood collection procedure consists of administering a questionnaire to screen for conditions that exclude participants from the blood draw. Fasting status is recorded. The urine collection procedure consists of urine specimen collection and processing.

The following exclusion criteria apply to all tests that require blood specimens:

- Hemophiliacs
- Participants who received chemotherapy within the last 4 weeks
- The presence of rashes, gauze dressings, casts, edema, paralysis, tubes, open sores or wounds, withered arms or limbs missing, damaged, sclerosed or occluded veins, allergies to cleansing reagents, burned or scarred tissue, shunt or intravenous lines on both arms.

Laboratory Quality Control and Monitoring

NYC HANES followed quality assurance and quality control (QA/QC) protocols that are used by NHANES and meet the 1988 Clinical Laboratory Improvement Act mandates. To ensure that the laboratory-reported values were accurate and reliable, standard quality control procedures were implemented at each of the testing facilities. These included the analysis of control specimens for which known values or concentrations have been previously assigned. Control specimens were included in each analytical run. Results from analytical runs where the control specimen readings were out of range were not reported. To monitor the reproducibility of results, testing was repeated on a 2% random sample of specimens. Periodic reports were generated by each of the testing laboratories to describe any out-of-range values, missing or inconsistent data, and control readings.

Detailed QA/QC instructions for the specific labs are discussed in the NHANES Laboratory/Medical Technologists Procedures Manual (LPM). Please refer to the NHANES General Documentation on Laboratory Procedures for detailed QA/QC protocols.

Description of Laboratory Methodology

Hepatitis C Antibody

- Antibodies to hepatitis C virus were measured at the New York City Public Health Laboratories. Serum samples were tested for anti-HCV with use of a second-generation enzyme immunoassay (EIA 2.0, Abbott Laboratories, North Chicago, IL). Samples that were positive were confirmed using Chiron RIBA HCV 3.0 Strip Immunoblot Assay (SIA).

Please see http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l02_c.pdf for a detailed description of the lab methodology.

Blood Lead, Cadmium and Mercury

- Blood specimens for metals, including lead, cadmium, and mercury, were tested at the New York State Wadsworth Laboratories using inductively coupled plasma mass spectroscopy (ICP–MS) methodology. Method detection limits for lead, cadmium, and mercury were 0.05 µg/L, 0.09 µg/L, and 0.17 µg/L, respectively. Please see http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l06bmt_c.pdf for a detailed description of the lab methodology.

Herpes Simplex Virus 1 and 2

- Serum samples were analyzed at Emory University School of Medicine for antibodies to HSV-1 and HSV-2 using type-specific immunodot assays. Purified glycoproteins gG-1 of HSV-1 and gG-2 of HSV-2 were used as antigens for HSV-1 and HSV-2 assays, respectively. These type-specific immunodot assays can differentiate between antibody to HSV-1 and HSV-2 with high sensitivity, specificity and reproducibility. Please see http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l09_c.pdf for a detailed description of the lab methodology.

Plasma Glucose and Glycohemoglobin

- Plasma glucose concentration and glycohemoglobin (HbA1C) were tested at the University of Missouri Diabetes Diagnostic Laboratory. Plasma glucose concentration was measured using an enzymatic reaction. HbA1C was measured on whole blood with an ion-exchange high-performance liquid chromatography (HPLC) method (Bio- Rad Diamat HPLC). Both of these methods were standardized to the Diabetes Control and Complications Trial reference method. Please see http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l10am_c.pdf and http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l10_c.pdf for detailed descriptions of the lab methodologies.

Lipid Profile

- For lipid profile measurements, blood samples were analyzed at the Lipoprotein Analytical Laboratory at Johns Hopkins University Hospital for fasting triglycerides, low density lipids (LDL), high density lipids (HDL) and total cholesterol. Serum cholesterol and triglycerides were measured enzymatically on a Hitachi 717 Analyzer (Boehringer Mannheim Diagnostics) using commercial reagents. Please see http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l13_c.pdf and http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l13am_c.pdf for detailed descriptions of the lab methodologies.

Serum Cotinine

- Serum cotinine levels were measured at the New York State Wadsworth Laboratories using an isotope dilution, liquid chromatography, tandem mass spectrometry method. The limit of detection (LOD) using this method was 0.050 ng/mL. The current NHANES method for serum cotinine was transferred and validated at New York State Wadsworth Laboratories after training in the author's laboratory at the National Center Environmental Health at CDC. However, the NHANES serum samples were analyzed using a newer, more sensitive mass spectrometer and thus had an LOD of 0.015 ng/mL.

- Briefly, the NYC HANES serum samples were equilibrated with a trideuterated cotinine internal standard and then extracted using pre-cleaned ChemElute solid phase extraction cartridges (Cat # 12198001 Varian, Palo Alto, CA), the extract evaporated to dryness under vacuum, reconstituted in 100ul of isopropanol and analyzed by LC/MS/MS using electrospray ionization. The instrumental system comprised an Agilent 1100 series LC and Applied Biosystems API 4000 triple quadrupole mass spectrometer. The limit of detection (LOD) for this method was 0.050 ng/mL cotinine in serum. Typical sample batches were 40 serum samples, at least 2 blanks, and high, medium, and low QC (15, 1.5 and 0.15 ng/ml respectively). All final results were blank corrected using the mean batch blank value. The average blank for the NYC HANES serum cotinine project conducted from 7/2004 to 2/2005 was 0.018 (n=440). Batch blanks were typically <0.03ng/ml throughout the analysis. Quality control charts for the three QC levels were evaluated to ensure data was only reported when the analysis was within control limits and that signals did not exceed the calibration range otherwise repeat analysis was performed.

Analytic Notes

Analytic Notes are available for some variables in LABS and are provided below by analyte. Analysts are encouraged to also refer to NHANES Data, Documentation, and Codebooks on specific sections by component for other notes and documentation (http://www.cdc.gov/nchs/about/major/nhanes/nhanes2003-2004/nhanes03_04.htm). Additionally, analysts may find that LAB variables should be utilized in conjunction with variables from other survey components (e.g., CAPI, ACASI).

'Previously Used Recodes' are provided for some measures if available. However, before using the recodes analysts must consider whether the recode is appropriate to their analysis and should review relevant literature and clinical guidelines for standard definitions of specific health outcomes. The list of 'Previously Used Recodes' is not exhaustive; the recodes are intended to help analysts understand how variables can be used in defining outcomes. Users may find that cutoffs or categories can be redefined using similar variables.

Analysts should refer to all materials under 'Using the Data' before analyzing the data. These materials provide guidance on how to use the data as well as the documentation available on the website. The Analytic Guidelines describe the specific weights in detail and when to use each one. The sample programs also illustrate examples for using each type of weight.

Hepatitis C Antibody

- Results of the screening EIA test (not released) were used in combination with the RIBA result (not released) to create a variable (**HCV**) that indicates the status of hepatitis C virus. This recoded variable **HCV** is released in the dataset and was determined as follows (http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l02_c.pdf):

    - Samples testing positive for anti-HCV by the screening EIA test were tested in the confirmatory RIBA assay for antibody to hepatitis C virus. Samples where the RIBA result was positive are reported as confirmed positive for antibody to HCV. Samples where the RIBA result was negative are reported as negative for antibody to HCV. Samples where the RIBA result was indeterminate are reported as indeterminate for antibody to HCV, however, analysts can consider reporting these as negative based on publications and NHANES documentation (http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l02_c.pdf).
    - Samples that tested negative by the screening EIA test were not tested by RIBA. These samples were reported as negative for antibody to HCV.

Plasma Glucose

- While plasma glucose was tested for all consenting survey participants who provided blood at the clinic, fasting plasma glucose measurements are available for a random sample of these participants (80%) who were assigned to fast for 8 hours prior to their appointment and prioritized for a morning interview. Persons self-identifying with a history of diabetes were not required to fast. Additional survey participants fasted voluntarily. Those who voluntarily fasted were compared to the SPs who were assigned to fast but did not. They were found to be similar on all demographic characteristics except age; therefore, they were included in the final analytic sample in lieu of the SPs who were assigned to fast but did not, and an age adjustment was made. Respondents for the fasting sample (n=1350) were SPs who fasted between 8 and 23 hours and either had a valid glucose reading or had ever been told by a health care professional that they had diabetes (**DIQ010** from CAPI). Analyses based on fasting plasma glucose should use the fasting weight (see the Analytic Guidelines).

- Based on published literature, diabetes estimates are generally presented as diagnosed diabetes and undiagnosed diabetes (See 'Previously Used Recodes' for diabetes):

    - Diagnosed diabetes, or self-reported diabetes, is based on participant report that a health care provider had ever told them that they had diabetes (other than during pregnancy for women) (**DIQ010**).
    - Participants without a prior diagnosis but whose fasting plasma glucose (**LBXGLU**) is greater than or equal to 126 mg/dL were considered to have undiagnosed diabetes.
    - Total diabetes is the combination of the measures for diagnosed and undiagnosed diabetes (i.e., participants who self-reported diabetes or whose fasting plasma glucose was greater than or equal to 126 mg/dL).

Lipid Profile

- Based on published literature, a commonly used definition of hypercholesterolemia incorporates total blood cholesterol lab measurements (**LBXTC**) with prescribed

cholesterol-lowering medication use (**BPQ100d** from CAPI). Hypercholesterolemia awareness, treatment and control can be estimated among individuals with hypercholesterolemia using **BPQ080, BPQ100d** and **LBXTC**. See 'Previously Used Recodes' for hypercholesterolemia.

- LDL-cholesterol is calculated from measured values of total cholesterol, triglycerides, and HDL-cholesterol according to the Friedewald calculation (see Previously Used Recodes for LDL):

  [LDL-cholesterol] = [total cholesterol] – [HDL-cholesterol] – [triglycerides/5]

  NOTES:
  - Triglycerides/5 is an estimate of very low-density lipoproteins (VLDL)-cholesterol.
  - All values are expressed in mg/dL.
  - The calculation is valid for triglycerides less than 400 mg/dL.

Serum Cotinine

- Participants with below LOD values were assigned a value of 0.035 ng/mL, which was based on the calculation LOD/square root of 2 (http://www.cdc.gov/exposurereport/pdf/results_02.pdf).

- For additional reporting requirements and information on interpreting levels of serum cotinine, please refer to the section on Results by Chemical Group for Tobacco Smoke in CDC's 2005 Third National Report on Human Exposure to Environmental Chemicals (http://www.cdc.gov/exposurereport/pdf/results_02.pdf).