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National Longitudinal Study of Adolescent to Adult Health (Add Health), 1994-2008 [Public Use]

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Wave IV: Biomarkers, Measures of Glucose Homeostasis Report

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2012

Add Health Wave IV Documentation

Measures of Glucose Homeostasis

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This research was supported by grant 5P01-HD31921 from the Eunice Kennedy Shriver National Institute of Child Health and Human Development, with cooperative funding from 23 other federal agencies and foundations. Further information may be obtained by contacting Add Health at addhealth@unc.edu.

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Add Health

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1. Introduction

During Wave IV, Add Health collected biological specimens from a large, nationally representative sample of young adults. Given the size of the Wave IV sample, its geographic distribution, and in-home setting of the respondent interviews, biological specimen collection involved practical, relatively non-invasive, cost-efficient and innovative methods. These methods included collection of capillary whole blood via finger prick by trained and certified field interviewers, its *in situ* desiccation, then shipment, assay and archival of dried blood spots. The collection of capillary whole blood followed the collection of cardiovascular and anthropometric measures (Entzel et al., 2009) and saliva (in preparation). It preceded the collection of data on respondent use of prescription and select over-the-counter medications (Tabor et al., 2010). Further details on the design of Add Health Waves I-IV, are available elsewhere (Harris, 2011).

Included in the Add Health Wave IV data are two measures of glucose homeostasis based on assay of the dried blood spots:

- Glucose (mg/dl) and
- Hemoglobin A_{1c} (HbA_{1c}, %)

To facilitate analysis and interpretation of HbA_{1c}, the restricted-use Add Health Wave IV data also include a trichotomous flag distinguishing the original (0) from two types (1,2) of inter-converted assay results (see Section 4.2.3.3):

- Convert (0,1,2)

Moreover, the restricted-use Add Health Wave IV data include six constructed measures:

- Fasting duration (h)
- Classification of fasting glucose (ADA, 2011)
- Classification of non-fasting glucose (ADA, 2011)
- Classification of HbA_{1c} (ADA, 2011)
- Anti-diabetic medication use
- Joint classification of glucose, HbA_{1c}, self-reported history of diabetes, and anti-diabetic medication use

This document summarizes the rationale, equipment, protocol, assay, internal quality control, data cleaning, external quality control, and classification procedures for each measure listed above. Documentation of other (metabolic; inflammatory; immune; genetic) measures based on

assay of the dried blood spots and genotyping of DNA extracted from salivary buccal cells will be provided in separate reports.

2. General Overview of Data Collection

A Blaise computer-assisted interview (CAI) program guided trained and certified field interviewers (FIs) through the blood spot collection process. Help screens with step-by-step measurement instructions were accessible within the program. Each FI also carried a Job Aids Booklet that served as a quick reference guide to study protocols.

Respondents were free to decline any or all measurements and specimen collections while participating in other components of the interview. In the Wave IV data set, any measures that are missing due to unique circumstances at correctional facilities are coded as legitimate skips.

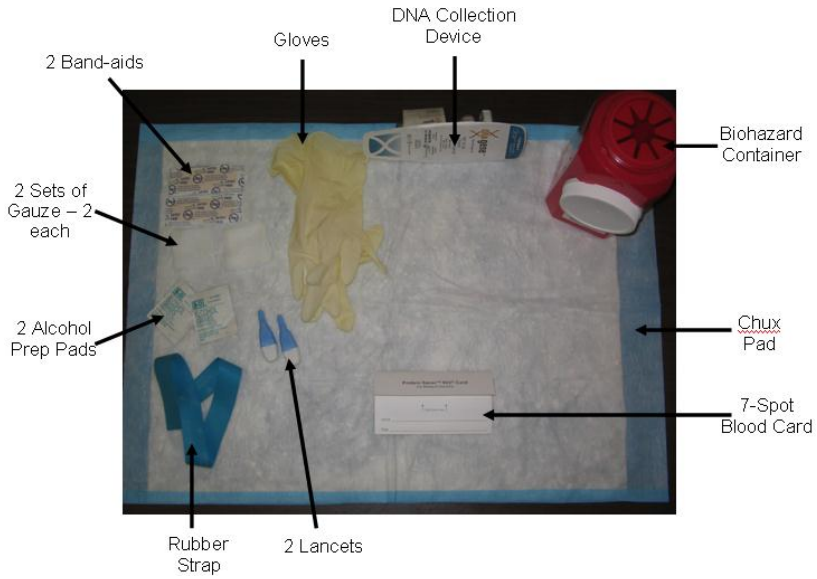
Some measurement protocols were revised in the period between the Wave IV Pretest (conducted in 2007) and the Main Study (conducted in 2008). Where the Pretest and Main Study data collection protocols differed significantly, this report documents the key differences between them. Pretest cases in the Wave IV data set are flagged for identification.

3. Capillary Whole Blood Collection

3.1 Rationale

Capillary whole blood was collected to provide Add Health with the biological specimens necessary to assay and interpret a pre-specified panel of metabolic, hematologic, inflammatory, and immune biomarkers, including the measures of glucose homeostasis described herein. It also was collected to establish a dried capillary whole blood spot archive capable of supporting future assays and ancillary studies.

Exhibit 1. Capillary whole blood collection equipment



3.2 Equipment

Sterile lancets, rubber strap, alcohol prep pads, gauze, Band-Aid type adhesive dressings, gloves, biohazard container, Chux-type absorbent underpad, and a seven-spot capillary whole blood collection card (Whatman 903® *Protein Saver*, Whatman Inc., Piscataway, NJ) to which a stabilizing, buffered preservative had been pre-applied to Spot #1 (*Exhibit 1*).

3.3 Protocol

3.3.1 Main Study

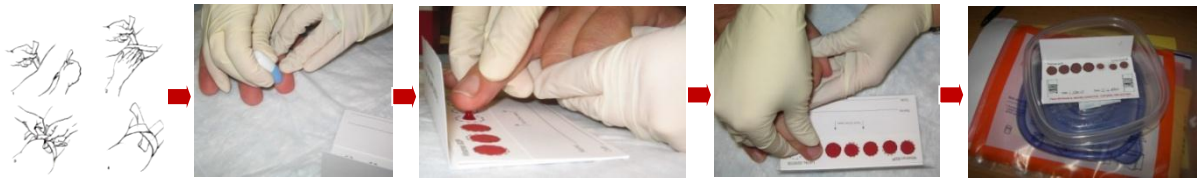
During the preceding anthropometric data collection procedure, all female respondents were asked specifically whether they had a prior mastectomy and, if so, on which side. If there were contraindications to using the right hand for capillary whole blood collection, the left hand was used. If there were contraindications on both hands, capillary whole blood was not collected.

FIs collected capillary whole blood from the respondent's middle or ring finger, unless one of the following contraindications was present:

- open sores, wounds, gauze dressings or rashes;
- casts, splints or shunts;
- intravenous (IV) catheters or other attached medical devices;
- swelling, withering or paralysis; or
- finger on same side as prior mastectomy.

FIs prepared the work surface for capillary whole blood collection and donned gloves. The help screen on the computer laptop directed FIs to refer to the Job Aids Booklet for the Fainting Protocol. FIs selected a finger for the procedure, cleaned it with the alcohol prep pad, and let it fully dry. While the finger was drying, FIs asked respondents to hang the selected finger below their waist while applying the rubber strap to the midpoint of the upper, ipsilateral arm. After placing the rubber strap, respondents started a timer on the laptop computer designed to sound an audible cue after three minutes to prompt removal of the rubber strap. FIs placed the clean finger against the work surface and firmly placed a sterile lancet against it to prick the fingertip, slightly lateral of center. FIs firmly wiped away the first drop of capillary whole blood with gauze, applying pressure to the base of (but not milking) the finger to facilitate flow. FIs were trained to allow a large droplet to accumulate before dropping it onto the first circle of the seven-spot capillary whole blood collection card and to do the same for the remaining six circles from left to right, all without allowing the fingertip to touch the card (*Exhibit 2*).

Exhibit 2. Collecting the capillary whole blood.



When seven capillary whole blood spots were successfully collected (or blood droplet formation ceased), FIs wiped off remaining blood with gauze, instructed respondents to firmly apply the gauze to the finger for at least two minutes, and then applied a band aid to it. FIs collecting fewer than five spots less than 80% full from a single prick requested respondents' permission to repeat the capillary whole blood collection procedure on a second finger from the contralateral hand. FIs asked respondents to discard used capillary whole blood collection equipment in their own trash receptacle (except for lancets which were discarded in the biohazard container). FIs discarded them in the biohazard container when interviews were conducted in public locations.

FIs bar code labeled each capillary whole blood spot collection card with the corresponding respondent biospecimen ID and then air dried it for three hours. Thereafter, FIs packaged each card with a desiccant pack and shipped it in a FedEx Priority Overnight envelope to the University of Washington Department of Laboratory Medicine (UW Lab Med, Mark H. Wener, M.D., Director, Seattle, WA) for assay.

UW Lab Med received the FedEx Priority Overnight envelopes containing a single dried blood spot collection card and desiccant pack. They scanned the FedEx tracking number and bar code-labeled card into a database in the order of receipt. They also keyed the receipt date, number of dried blood spots per card (0-7), number of adequate blood spots per card defined by blood filling ≥ 80% of the target area (0-7), comments on dried blood spot quality, and condition of the desiccant pack alongside the biospecimen ID. They grouped the cards (≤ 25 per group), sealed the groups in Ziploc bags with desiccant packs, and stored them at -70°C until processing. Immediately before processing, they warmed cards to room temperature (23°C) and re-scanned the bar code-labeled card into the database. The cards were punched for all assays except HbA_{1c}, returned to the freezers, then shipped frozen to the Carolina Population Center (CPC, University of North Carolina, Chapel Hill, NC) for permanent archival. At the CPC, Spot #1 on each card was removed, re-bundled (≤ 25 per group), sealed in plastic bags with desiccant, and shipped frozen by next day air to FlexSite Diagnostics, Inc. (Robert A. Ray, Ph.D., Director, Palm City, FL) for HbA_{1c} assay.

3.3.2 Pretest Methodological Variations

During the Pretest, respondents chose the middle or ring finger and FIs were directed not to use the thumb, index finger or fifth digit/little finger for capillary whole blood collection. FIs also collected up to ten capillary whole blood spots: three on a BIOSAFE Blood Collection Card for HbA_{1c} and Cholesterol Panel (BIOSAFE Laboratories, Inc., Chicago, IL) and seven more on a Whatman 903® Protein Saver, (Whatman Inc., Piscataway, NJ). The BIOSAFE card was made of Whatman 903® filter paper (Whatman International, Dassel, Germany) with a top layer of TELFA (Kendall Healthcare Products, Mansfield, MA) to minimize the effects of blood spot layering and inadvertent touching of cards with respondent fingertips (Tyrrell, 1999; Maggiore, 2002; Bui et al., 2002a; Bui et al., 2002b; Grzeda et al., 2002). A stabilizing borate buffered preservative was pre-applied by BIOSAFE to the area of the BIOSAFE card designated for HbA_{1c} assay and dried. As in the main study, the Whatman 903® Protein Saver card was shipped to UW Lab Med for assay.

4. Measures of Glucose Homeostasis

4.1 Glucose

4.1.1 Rationale

Glucose was assayed in dried capillary whole blood spots because of its central role in the identification of impaired fasting glucose / pre-diabetes, and diabetes mellitus, two well-known risk factors for cardiovascular disease morbidity and mortality.

4.1.2 Assay and Internal Quality Control

UW Lab Med constructed dried blood spot glucose assay calibrators from pooled human plasma spiked with a concentrated glucose solution (Sigma, St. Louis, MO) and serially diluted with 7% bovine serum albumin in phosphate buffered saline (BSA/PBS; Sigma) to the desired final glucose concentration. Two dried blood spot quality control samples were constructed from a separate pool of human plasma, either undiluted (high glucose concentration quality control sample) or diluted with BSA/PBS (low glucose concentration quality control sample). Each calibrator and quality control sample solution was mixed 1:2 with washed human erythrocytes (UW Lab Med), pipetted in 75µl aliquots onto #903 filter paper (Whatman Inc., Piscataway, NJ) and dried for 4 hr at room temperature. The glucose concentration of each calibrator and quality control sample solution was determined by analysis on a Synchron LX 20 Pro Clinical System (Beckman Coulter, Miami, FL). Dried blood spot calibrators and quality control samples were stored at -70°C in sealed Ziploc bags with desiccant until use.

A single 3.2 mm (1/8 in) diameter punch was taken from each dried blood spot calibrator, quality control sample or respondent sample and placed into a deep-well microtiter plate well (Greiner Bio-One, Monroe, NC). Plates were either immediately assayed or were firmly sealed and stored

at -70°C pending assay. Plates to be assayed were warmed to room temperature and then 75 µl of elution solution (PBS; Sigma) was added column-by-column to each well using an eight-channel electronic pipettor (Rainin, Oakland, CA). Plates were sealed and vigorously shaken for 1 hr on a Delfia Plateshake microplate shaker (PerkinElmer, Waltham, MA). A 190 µl aliquot of glucose reagent (Synermed, Westfield, IN) was added column-by-column to the elution solution in each well, the plate sealed and the resulting reaction mixture incubated at 37°C for 30 min. A 190 µl aliquot of the reaction mixture was transferred column-by-column from each well to a flat-bottom, shallow-well microtiter plate (Greiner Bio-One, Monroe, NC). The optical density of each well (in arbitrary units) was then read at 660 nm on a Synergy HT Microtiter Plate Reader (BioTek, Winooski, VT). Plate-specific linear regression calibration curves, electronically constructed by plotting the concentrations of the calibrators against absorbance values, were used to convert the optical density of each respondent sample into a dried blood spot glucose concentration (BioTek Gen5). Respondent samples with a glucose concentration greater than the highest calibrator were diluted and re-assayed. The sensitivity of the glucose assay was 22 mg/dl, the within-assay coefficient of variation was 4.4% and the between-assay coefficient of variation was 4.8%. Glucose concentrations (mg/dl) of 83 paired dried blood spot and serum samples determined using the protocol described above and the conventional Beckman Coulter UniCel DxC 800 Synchron Clinical System were strongly correlated (Pearson r = 0.97) and linearly associated in Deming regression (Cornbleet et al., 1979; Deal et al., 2011): serum glucose (mg/dl) = 1.193 * dried blood spot glucose (mg/dl) + 2.617. Correction for hematocrit did not increase the strength of association.

4.1.3 Pretest Methodological Variations

None.

4.2. Hemoglobin (HbA_{1c})

4.2.1 Rationale

HbA_{1c} was assayed in dried capillary whole blood spots because it is an integrated measure of glucose homeostasis, reflecting average blood glucose over the preceding two to three months. The measure plays a critical role in the management of diabetes since it is correlated with micro- and macrovascular complications and is widely used as the standard biomarker for the adequacy of glycemic management (American Diabetes Association, 2011).

4.2.2 Assay and Internal Quality Control - Flexsite 1 HbA_{1c} Assay

FlexSite Diagnostics placed a 6.35 mm (1/4 in) diameter punch from each dried blood spot respondent sample in a microtiter plate, eluted blood from the punches, released hemoglobin (Hb) from the blood via hemolysis, proteolytically degraded it with pepsin, and oxidized heme

groups using Roche Unimate reagents. Hb and HbA_{1c} were then measured in the hemolysate on the Roche COBAS INTEGRA® 700 Analyzer (Roche Diagnostics, Indianapolis, IN) (Roche Diagnostics, 2005). Total Hb was measured using a cyanide-free colorimetric method based on the formation of a brownish-green chromophore, alkaline hematin D-575, in alkaline detergent solution (Zander et al., 1984). The color intensity (proportional to the total Hb concentration in the respondent sample) was determined by monitoring the increase in absorbance at 552 nm. Total Hb was calculated using a fixed factor determined from the primary calibrator, chlorohemin (Wolf et al., 1984). HbA_{1c} was measured using monoclonal antibodies attached to latex particles. The antibodies bound the β-N-terminal fragments of HbA_{1c}. Remaining free antibodies were agglutinated with a synthetic polymer carrying multiple copies of the β-N-terminal structure of HbA_{1c}. The change in turbidity (inversely related to the amount of bound glycopeptide in the respondent sample) was measured turbidimetrically at 552 nm. A synthetic polypeptide compromising the N-terminal structure of HbA_{1c} was used for calibration (Little et al., 1992). HbA_{1c} (%) was calculated from the HbA_{1c}:Hb ratio, as follows: HbA_{1c} (%) = 2.27 + 87.6 × (HbA_{1c} ÷ Hb) (Rohlfing et al., 2002; NGSP, 2011). At a total Hb of 13.2 g/dl, the sensitivity of the conventional Roche HbA_{1c} assay was 3%. The within- and between-run coefficients of variation assay were 2.2%-2.4% (Roche Diagnostics, 2005). HbA_{1c} (%) of 80 paired dried blood spot and whole blood samples determined using the protocol described above and the conventional HbA_{1c} assay on the Tosoh SRL#5 (Tosoh Bioscience, Inc., South San Francisco, CA) were strongly associated (Pearson r = 0.99) and linearly related: dried blood spot HbA_{1c} (%) = 1.014 × whole blood HbA_{1c} (%) – 0.296.

4.2.3 Pretest Methodological Variations

During the Pretest period, HbA_{1c} was measured by BIOSAFE Laboratories, Inc. in 53 respondents and by FlexSite Diagnostics, Inc. in 129 respondents using a slightly modified version of the Roche Unimate / COBAS INTEGRA HbA_{1c} assay described above. Descriptions of the assays and the inter-conversion of their results follows.

4.2.3.1 Assay and Internal Quality Control - Flexsite 2 HbA_{1c} Assay

The Roche Unimate / COBAS INTEGRA HbA_{1c} assay described above was changed to a Roche Tina-quant Gen 2 / COBAS INTEGRA HbA_{1c} assay to reduce potential interference from hemoglobin variants such as HbS, found in sickle cell trait (NGSP, 2011).

4.2.3.2 Assay and Internal Quality Control - BIOSAFE HbA_{1c} Assay

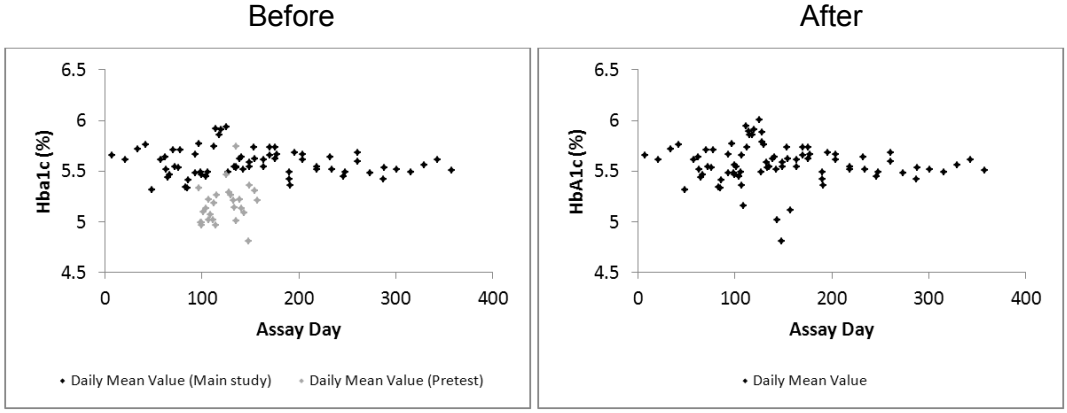
Two, 3.0 mm (1/8 in) diameter punches were collected from a homogeneous region of each dried blood spot respondent sample using a calibrated hole punch (McGill Inc., Marengo, IL). The punches were transferred to a pre-labeled 15 x 100 mm borosilicate tube (Fisher Scientific,

Pittsburgh, PA). To each tube, 630 µl of Roche Hemolyzing Reagent (Roche Diagnostics, Indianapolis, IN) were added. The tubes were rotated at 1000 rpm for 60 min at 25°C. Following centrifugation at 2400 rpm (1300g) at 25°C for 10 min, the eluate was analyzed for HbA_{1c} using the Roche P-Modular System (Roche Diagnostics, Indianapolis, IN) and Roche Hemoglobin A_{1c} Reagents which employ an immunoturbidimetric method for HbA_{1c} quantitation and a colorimetric method for Hb quantitation, as described above. HbA_{1c} (%) was calculated using the formula: HbA_{1c} (%) = 91.5 × (HbA_{1c} ÷ Hb) + 2.15 (Roche Diagnostics, 2006). The Roche HbA_{1c} method was certified by the National Glycohemoglobin Standardization Program as being traceable to the HbA_{1c} reference method (NGSP, 2011). Analytical runs were quality controlled with commercially available assayed whole blood control materials (BioRad Laboratories, Hercules, CA), and laboratory-prepared dried blood quality control cards (Streck, Inc., Omaha, NE). At HbA_{1c} between 5.0% and 11.0%, the ranges of within- and between-run coefficients of variation for the BIOSAFE HbA_{1c} assay were 1.4%-3.2%, and 3.5%-7.6%, respectively. HbA_{1c} (%) of 115 paired dried blood spot and whole blood samples determined using the protocol described above and a conventional HbA_{1c} assay were strongly associated (Pearson r = 0.99) and linearly related: dried blood spot HbA_{1c} (%) = 0.850 × whole blood HbA_{1c} (%) + 0.8079 (BIOSAFE, 2006).

4.2.3.3 Inter-Conversion of FlexSite 1, FlexSite 2 and BIOSAFE HbA_{1c} Assay Results

Four Add Health Wave IV Main Study respondents were randomly selected from those (n = 14,148) with FlexSite 1 HbA_{1c} assay results within each of 24 strata defined by race / ethnicity (non-Hispanic white; non-Hispanic black; Hispanic; other), sex (male; female) and HbA_{1c} tertile (low, medium, high). Collectively, the stratified, random sample included 4 × 24 = 96 respondents whose dried capillary whole blood spot collection cards were resubmitted for Flexsite 2 HbA_{1c} assay by laboratory staff masked to respondent identity. The association between the natural log transformed FlexSite 1 and FlexSite 2 HbA_{1c} assay results was modeled using Deming regression (Cornbleet et al., 1979; Deal et al., 2011) , i.e. log(FlexSite 1) = α + β × log(FlexSite 2) (Pearson r = 0.63). Estimates of α, β and their 95% confidence intervals (CIs) were 0.22 (-0.13, 0.58) and 0.85 (0.65 to 1.06). α and β were used to convert FlexSite 2 to FlexSite 1 HbA_{1c} assay results among (n = 129) Pretest respondents with missing FlexSite 1

Exhibit 3. Mean HbA_{1c} (%), before & after inter-conversion of assay results



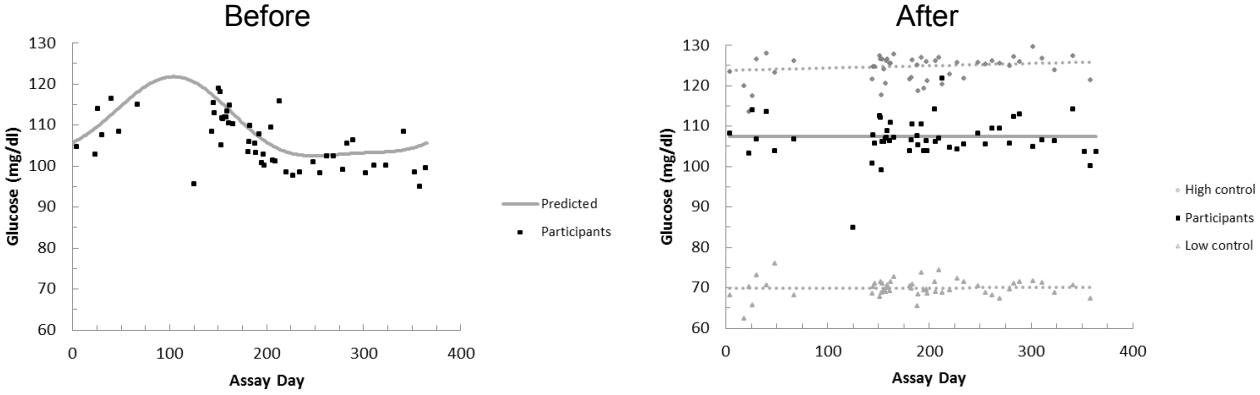
assay results (Convert=1). The association between the natural log transformed FlexSite 2 and BIOSAFE HbA_{1c} assay results also was modeled using Deming regression (Cornbleet et al., 1979; Deal et al. 2011) , i.e. log(FlexSite 2) = $\gamma + \delta \times$ log(BIOSAFE) (Pearson r = 0.66). Estimates of γ, δ and their 95% CIs were -1.91 (-3.20, -0.61) and 2.22 (1.43, 3.02). α, β, γ and δ were used to serially convert BIOSAFE to FlexSite 2 to FlexSite 1 HbA_{1c} assay results among (n = 53) Pretest respondents with missing FlexSite results (Convert=2). Inter-conversion of HbA_{1c} attenuated among-assay differences (*Exhibit 3*).

5. Data Cleaning and External Quality Control

5.1 Data Cleaning

Plate-specific, linear regression calibration formulae (*Section 4.1.2*) were used to verify laboratory conversion of optical density to glucose concentration (mg/dl). Dried blood spot respondent samples with glucose (mg/dl) in the lowest half percentile of the distribution were re-assayed. Original and repeat assay values were averaged. Seasonal variation was examined by plotting dried blood spot and quality control glucose (mg/dl) versus assay date (2007-2009) on a single calendar time scale (1-366 days). A previously described pattern of seasonal variation (Gikas et al., 2009; Kershenbaum et al., 2011; Tseng et al., 2005) was observed (*Exhibit 4*). Glucose (mg/dl) was therefore modeled as a function of time, represented by four seasonal terms: sine(2Πjt÷366) and cosine(2Πjt÷366), where harmonic j = (1, 2) and time t = (1, 2, 3, …, 366). This non-linear model was adjusted for age (yr), sex (male; female), and race/ethnicity (non-Hispanic white; non-Hispanic black; non-Hispanic Asian/Pacific Islander; Mexican; other Hispanic/Latino; other race/multiracial). Goodness of fit as measured by the Akaike information criterion was greatest for this model when compared to a variety of non-linear and linear alternatives. The age-, sex- and race/ethnicity-adjusted harmonic terms estimated in the former model were subtracted from the assayed values of glucose (mg/dl). Subtraction had little effect on the overall distribution, mean, or variance of glucose, but attenuated the observed seasonal variation (*Exhibit 4*).

Exhibit 4. Mean glucose (mg/dl), before and after decycling assay results



5.2 External Quality Control

Within a race / ethnicity- and sex-stratified random sample of (n = 100) Add Health respondents among whom capillary whole blood was collected twice, one-two weeks apart, reliabilities of random (*fasting or non-fasting*) glucose (mg/dl) and interconverted HbA_{1c} (%) were estimated as intra-class correlation coefficients (95% CI): 0.40 (0.22, 0.59) and 0.97 (0.96-0.98), respectively. The reliability of (*fasting*) glucose was 0.67 (0.35-1.00) within the fasting subset of these respondents (n = 23). Among three dried blood spot quality control samples assayed by masked FlexSite staff twice weekly for ten weeks (n = 60), mean absolute and relative bias (standard deviation) of interconverted HbA_{1c} were 0.2% (0.1%) and 3.8% (2.8%) when compared to HbA_{1c} conventionally assayed in paired whole blood on the Tosoh G7 Automated HPLC-HbA_{1c} Analyzer (Tosoh Bioscience, Inc., South San Francisco, CA) by Duke University Health System Laboratories (Durham, NC). HbA_{1c} (%) was strongly correlated with fasting glucose (mg/dl) (Pearson r = 0.68). The HbA_{1c}-based estimated average glucose (Nathan et al., 2008) and mean plasma glucose (Rohlfing et al., 2002) were 8% and 16% higher than fasting glucose (mg/dl). Measures of the HbA_{1c}-fasting glucose correlation and HbA_{1c}-based overestimation of fasting glucose were similar in the National Health and Nutrition Examination Survey (2007-2008): Pearson r = 0.75, 7% and 12%, respectively. These findings suggest that carefully standardized, in-home collection of whole blood spots can yield valid and reliable measures of glucose homeostasis (Whitsel et al., 2012).

6. Constructed Measures

6.1 Fasting Status

Variable: FASTTIME

In Interview Section 4: General Health and Diet, field interviewers asked respondents the following question: "*At what time did you last eat or drink anything other than water, including sugar-containing candy or gum?"* The response to the question was recorded in hours and minutes and designated AM or PM. On-screen instructions of "12 MIDNIGHT IS AM. 12 NOON IS PM" were provided to the field interviewer for clarification. To calculate fasting times, responses to the above question were subtracted from the laptop timestamp at the start of the interview. Since laptop clocks were set to Eastern Time for field work processing, the CPC Spatial Analysis Unit corrected interview start times using geocoded respondent locations, the time zones in which they were located, and the U.S. Daylight Savings Time schedule. In a final step, fasting times also were adjusted for interviews that stopped prematurely, but resumed later. Data on fasting times (range: 0-23.9 hr) were missing for 141 respondents, the result of recoding implausible values (> 24 hr), missing time zone data, breakoff interviews that resumed before 24 hours elapsed such that the question was not repeated, unrealistic computer timestamps, and answers of "*don't know*" or "*refused*" to the question.

6.2 Classification of Fasting Glucose

Variable: C_FGLU

The classification of glucose concentrations among Add Health respondents who were fasting (≥ 8 hr) at the time of blood collection was constructed based on the 2011 clinical practice recommendations for the diagnosis and classification of diabetes (ADA, 2011). Classes of fasting glucose are defined as follows:

Fasting* Glucose (mg/dl)	ADA Class
100-125	Impaired Fasting Glucose (IFG) / Pre-Diabetes
≥ 126	Diabetes

*Fasting for ≥ 8 hr

6.3 Classification of Non-Fasting Glucose

Variable: C_NFGLU

The classification of glucose concentrations among Add Health respondents who were non-fasting (< 8 hr) at the time of blood collection was constructed based on the 2011 clinical practice recommendations for the diagnosis and classification of diabetes (ADA, 2011). Classes of non-fasting glucose are defined as follows:

Non-Fasting* Glucose (mg/dl)	ADA Class
≥ 200	Diabetes

*Fasting for < 8 hr

6.4 Classification of HbA_{1c}

Variable: C_HBA1C

The classification of HbA_{1c} among Add Health respondents was constructed without regard to fasting status at the time of blood collection based on the 2011 clinical practice recommendations for the diagnosis and classification of diabetes (ADA, 2011). Classes of HbA_{1c} are defined as follows:

HbA_{1c} (%)	ADA Class
5.7-6.4	Pre-Diabetes
≥ 6.5	Diabetes

6.5 Anti-diabetic Medication Use

Variable: C_MED

Respondents who report using medications in the past four weeks associated with one or more of the following therapeutic classification codes

(99) Antidiabetic Agents,
(213) Sulfonylureas,
(214) Non-Sulfonylureas,
(215) Insulin,
(216) Alpha-Glucosidase Inhibitors,
(271) Thiazolidinediones,
(282) Meglitinides,
(309) Miscellaneous Antidiabetic Agents,
(314) Antidiabetic Combinations,
(371) Dipeptidyl Peptidase 4 Inhibitors,
(372) Amylin Analogs,
(373) Incretin Mimetics

were assigned a value of 1 for C_MED, anti-diabetic medication use. Those who did not report taking medications in these classifications were assigned a value of 0. (Tabor et al., 2010)

6.6 Joint Classification of Glucose, HbA_{1c}, Self-Reported History of Diabetes, and Anti-Diabetic Medication Use

Variable: C_JOINT

Add Health respondents also were classified as having diabetes if they had a fasting glucose ≥ 126 mg/dl (C_FGLU = 3) or non-fasting glucose ≥ 200 mg/dl (C_NFGLU = 2) or $HbA_{1c} \geq 6.5\%$ (C_HBA1C = 3) or self-reported history of diabetes except during pregnancy (H4ID5D = 1) or used anti-diabetic medication (C_MED=1) in the past four weeks.

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