

Vitamin D Biomarker Analytical Issues in NHANES



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**Centers for Disease Control and Prevention
National Center for Health Statistics**



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Presentation Objectives

- n Provide brief background on NHANES and Vitamin D biomarker data
- n Discuss NHANES serum 25(OH)D assay challenges and recent data advisory note
- n Discuss recent Roundtable meeting
- n Discuss next steps by NCHS and federal partners relative to Vitamin D biomarker



What is NHANES?

- n National Health and Nutrition Examination Survey
- n Conducted by the National Center for Health Statistics (NCHS) of the CDC
- n To assess the health and nutritional status of a nationally representative sample of US population
- n Data used for a variety of research and programmatic purposes



What is the Vitamin D status of the US Population?

- n The principal biomarker of vitamin D status is serum 25-hydroxyvitamin D or 25(OH)D.
- n 25(OH)D data collected in many National Health and Nutrition Examination (NHANES) Surveys.
- n 25(OH)D data: NHANES III (1988-94); NHANES 2000-2006 and 2007 and beyond



NHANES: Serum 25OHD Samples

<i>Survey</i>	<i>Ages</i>	<i>Sample</i>
NHANES III (1988-1994)	12+	18,158 ^a
NHANES 2000 (not on public use file)	6+	4,188 ^a
NHANES 2001-02	6+	7,807 ^{a,b}
NHANES 2003-04	1+	8,294 ^a
NHANES 2005-06	1+	7,402 ^b

^aLooker et al., 2007 NIH conference; ^bPfeiffer, Roundtable handouts, 2009



NHANES: Serum 25OHD Assays

n Laboratory:

- National Center for Environmental Health (NCEH), CDC

n Methods:

- DiaSorin RIA (25OHD₂ + 25OHD₃)
 - n 1988-94 and 2000-2006
- LC-MS/MS
 - n 2007 and after

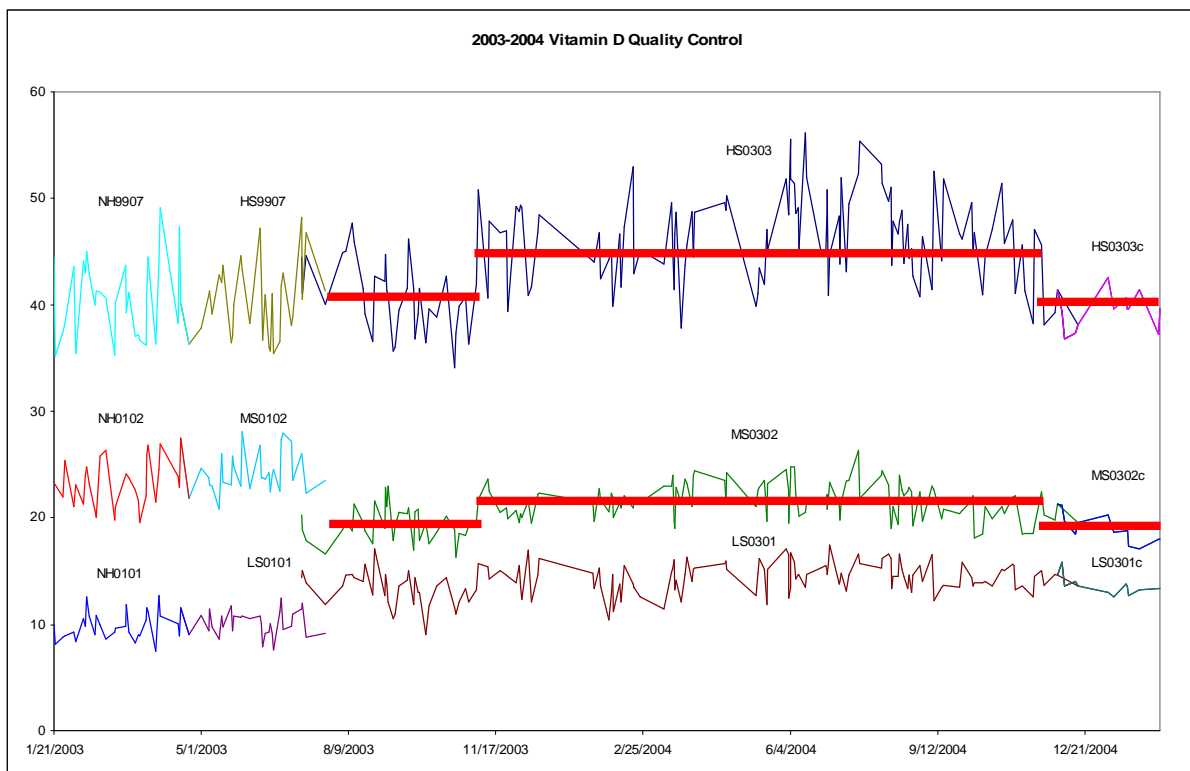


Initial 25(OH)D Assay Concerns (Looker et al., 2008)

- n 1988-94 vs. 2000 - 2004:
 - Reformulation (introduction of antibody to improve binding) of Diasorin RIA assay kit → shifts in assay results between these two time periods
- n 2000-2004:
 - Likely affected by drifts in the assay performance (method bias and imprecision) over time
- n Adjusted for assay drifts to compare time trends in 25(OH)D and identify contributing factors

Review of 2004 QC performance

- DiaSorin RIA assay was performing on the high side of the QC limits during 2004, particularly for the high pool
 - Late 2003 – new lot number for standards, tracer, other reagents
 - Late 2004 – new lot number of tracer, antiserum
- Conclusion: Conduct a 150 sample crossover study for QC shift



3. 2004 assay shift



±Adjustment Affects Conclusions

- n Adjustments for assay differences:
 - .. Looker et al. (2008):
 - n Overall mean 25(OH)D lower in 2000-2004 than in 1988-94
 - n Assay changes accounted for much of the difference
- n No adjustment for assay differences:
 - .. Ginde et al. (2009): Marked decrease in 25(OH)D in 2000-2004 compared to 1988-1994
 - n Growing epidemic of vitamin D insufficiency
 - .. Saintonge 2009: Vitamin D deficiency is increasing from 1988-1994 to 2006
 - n Implementation of national fortification and public health strategy



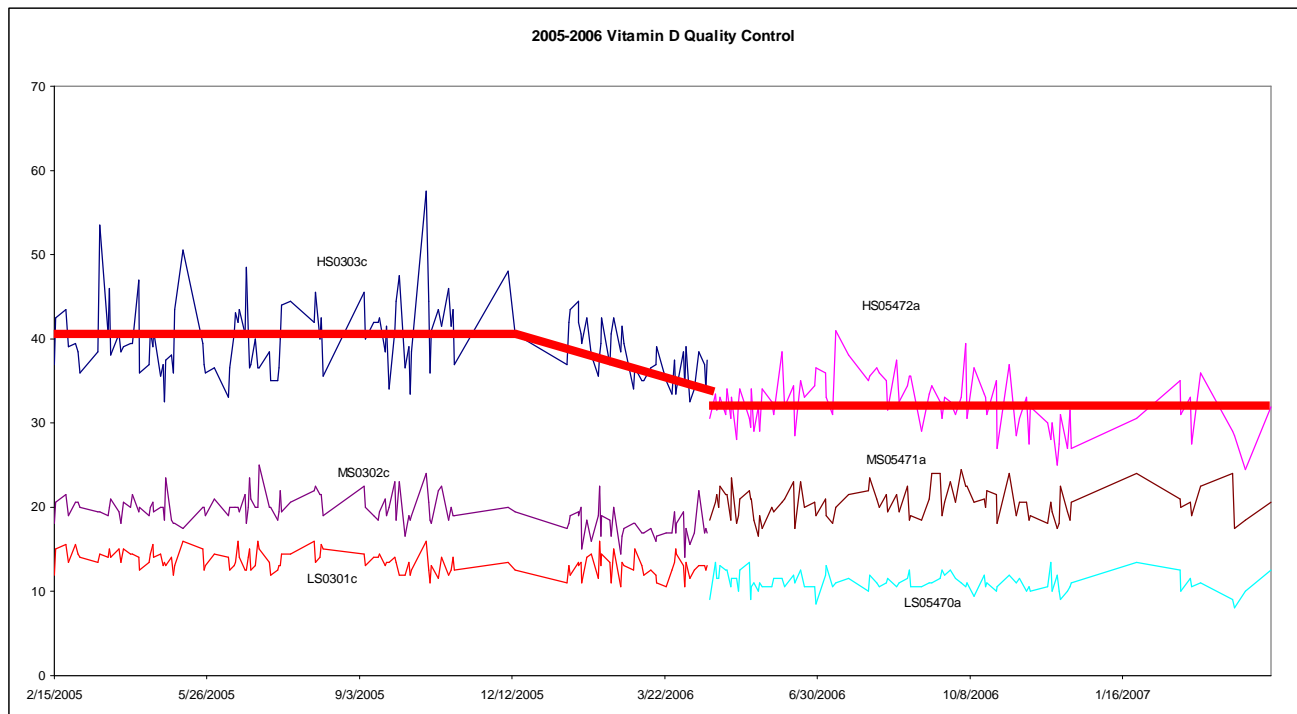
NCHS Analytic Note (2009)

- n Published data 2000 – 2004:
 - .. Effect of drifts in assay comparability for the serum 25(OH)D data collected in this time period may not be fully accounted for
- n Additional drifts in assay comparability for the 2005 – 2006 results
- n Cautions users when assessing time trends (especially using 2-year data sets)

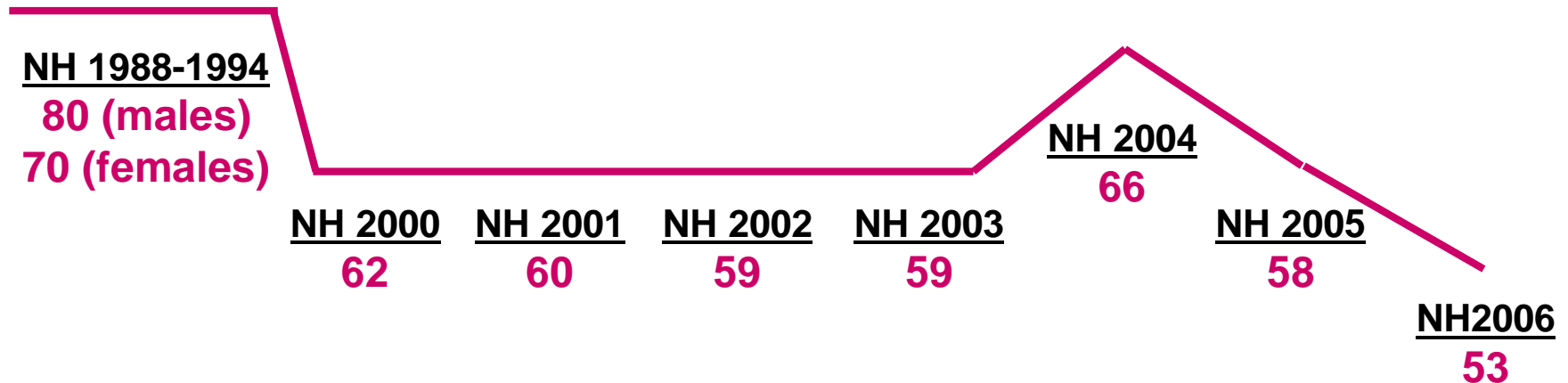
http://www.cdc.gov/nchs/data/nhanes/nhanes3/VitaminD_analyticnote.pdf

Review of 2006 QC performance

- DiaSorin RIA assay was performing on the low side of the QC limits during 2006, particularly for the high pool
 - Early 2006 – new lot number for tracer, antiserum and other reagents
 - End date? Stopped using 0301/0302/0303 series QC in Spring 2006; new 05470/05471/05472 series QC (characterized during Spring 2006) performed on the same level from Spring 2006 through Spring 2007



25OHD (nmol/L) NHANES weighted means track well with trends seen in QC data



For methods comparison only and not for publication. Not adjusted for reformulation or assay shifts



Multi-year estimates of 25OHD (nmol/L)

Survey years	Public data	n	Mean	Percentile			Percent	
				5th	50th	95th	<37.5	<75
2001-2002	Y	7,807	60 (58-61)	23	57	97	16 (13-19)	77 (73-80)
2003-2004	Y	7,399	63 (59-66)	24	60	103	13 (10-18)	71 (66-76)
2005-2006	Y	7,402	55 (53-57)	20	53	90	20 (17-24)	83 (80-85)
2001-2004	Y	15,206	61 (59-63)	24	59	101	15 (12-17)	74 (71-77)
2003-2006*	Y	14,801	59 (57-61)	22	56	97	17 (14-20)	77 (74-80)
2001-2006*	Y	22,608	59 (58-61)	22	56	97	16 (15-19)	77 (74-79)
2000-2006*	N	26,796	59	23	57	97	16	76

* "Odd" QC years cancel out





Roundtable (July 27-28, 2009)

- n Jointly sponsored by National Center for Health Statistics (CDC) and the Office of Dietary Supplements (NIH)
- n Discuss 25(OH)D assay challenges and options for resolving them
- n Biochemists, clinical laboratorians and biostatisticians



Roundtable Discussions

- n Laboratory QC data
- n Options for harmonization across Diasorin-based surveys (1988-1994 and 2000-2006)
- n Plans for NHANES 2007-2008 and beyond



NHANES 25(OH)D: Next Steps

- n Today: Data Advisory posted on website
- n Near term: Explore adjustment procedures to account for assay drift and reformulation and update data advisory
- n Longer term: Utilize NIST standard reference material to bridge past and future 25(OH)D data collected in NHANES



<http://www.cdc.gov/nchs/nhanes.htm>