

SWAN Repository Dataset Codebook

Study: App001_R37, "Estrogen Metabolism, Diet and Smoking", MFSowers

Dataset name: SWANRep001R37_Isoprostanes

Cohort: SWAN, all sites

The "isoprostane" dataset contains the isoprostane results of 1709 SWAN women who had an annual visit urine sample at V05 (The DHS samples were used for 44 site 13 women). Isoprostanes were measured as part of a study on isoprostanes and E2 in 2006. The CSAP of this study is attached in a separate document.

In the "isoprostane" dataset, the vast majority (N=1676) of isoprostane values fell within 39 to 5000 pg/mL, and these values are reported in the "isoprostane" column. Six isoprostane values were below 39 pg/mL; in the "isoprostane" column, these six observations are set to be "40.3". Finally, 27 isoprostane observations were above 5000 pg/mL. In the "isoprostane" column, these observations were replaced with random numbers generated according to this algorithm:

1. First, create a random variable x . x has 27 observations and each observation was a random number drawn from a uniform distribution between 0 and 1.

2. Second, calculate y from x according to the following rules:

if $x < 0.333$	then $y = x$
if $0.333 \leq x < 0.667$	then $y = x + 0.5$
if $x \geq 0.667$	then $y = x - 0.5$

3. Third, calculate the values to be used to replace those extreme values according to the following rules:

first 9 observations:	$\text{new_isoprostane} = y * 100 + 4350$
next 9 observations:	$\text{new_isoprostane} = y * 100 + 4550$
last 9 observations:	$\text{new_isoprostane} = y * 100 + 4750$

4. Fourth, sort those isoprostane observations that need "imputation" by id and sort the dataset containing "new_isoprostane" by the value of "y" (from small to large).

5. Merge and replace the extreme isoprostane values with "new_isoprostane". The result was smaller IDs taking on "new_isoprostane" values calculated from smaller y 's.

The isoprostane replacement values are decisions from previous analysis. Site 11 has a copy of isoprostane raw data without replacement values.

Dataset contents

Variable	Label	Comments
Sequence_no	Sequence_no	A sequential observation identifier
CVpct	CVpct	% Coefficient of variation of repeated measurements on the same sample. Used by the lab for quality control purposes.
Dilution	Dilution	Dilution factor
F2alsoprostane	F2alsoprostane	Name of the analyte. In this case, it's all isoprostane.
isoprostane	Adj_Result	Isoprostane results. These results have accounted for the dilution factor. In addition, 33 isoprostane values have been replaced using numbers/algorithm described above.
Unit	Unit	Unit of isoprostane
Accepted	Accepted	Whether the quality of isoprostane data was acceptable
Assay	Assay	Name of the assay
matrix	matrix	Matrix of the analyte. In this case, it's all urine.
Hi_Lowflag	Hi_Lowflag	A flag variable indicating whether isoprostane was "<39", "ok" or ">5000.0".
ARCHID		Encrypted SWAN Subject ID
VisitNum		SWAN visit number.
randflag	Isoprostane value flag. 0:Actual value. 1:Value replaced with random number. 2:Value set to be 40.3.	A flag variable indicating whether the result reported in the "isoprostane" column was "an actual measured value", "a value replaced with random number" or "a value set to be 40.3".

F2 Isoprostane assay description

[iPF_{2α}-III EIA Kit; 8-epi Prostaglandin F_{2α} EIA Kit; 8-Isoprostane ELISA Kit; 8-iso Prostaglandin F_{2α} EIA]

This Cayman assay is based on the competition between 8-isoprostane and an 8-isoprostane-Acetylcholinesterase (AChE) conjugate (8-Isoprostane Tracer) for a limited number of 8-isoprostane-specific rabbit antiserum binding sites. Because the concentration of the 8-Isoprostane Tracer is held constant while the concentration of 8-isoprostane varies, the amount of 8-Isoprostane Tracer that is able to bind to the rabbit antiserum will be inversely proportional to the concentration of 8-isoprostane in the well. This rabbit antiserum-8-isoprostane (either free or tracer) complex binds to the rabbit IgG mouse monoclonal antibody that has been previously attached to the well. The plate is washed to remove any unbound reagents and then Ellman's Reagent (which contains the substrate to AChE) is added to the well. The product of this enzymatic reaction has a distinct yellow color and absorbs strongly at 412 nm. The intensity of this color, determined spectrophotometrically, is proportional to the amount of 8-Isoprostane Tracer bound to the well, which is inversely proportional to the amount of free 8-isoprostane present in the well during the incubation.

Plasma, serum, urine, whole blood, as well as other heterogeneous mixtures can interfere in the assay. It is best to check for interference before embarking on a large number of sample measurements. To test for interference, dilute one or two test samples to obtain at least two different dilutions of each sample between 5 and 200 pg/ml (i.e., 20-80% B/B₀). The two different dilutions of the sample show good correlation (differ by 20% or less) in the final calculated 8-isoprostane concentration, purification is not required. If you do not see good correlation of the different dilutions, purification is advised. Cayman offers an 8-Isoprostane Affinity Column and Affinity Sorbent are recommended as the easiest and most convenient purification format for 8-isoprostane. The affinity column purification procedures have been validated with plasma and urine samples.

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