**Appendix 1**

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In the National Health and Nutrition Survey 2012 (ENSANUT-2012), we observed an 11.25 mg/dl decrease in the concentration of glucose in comparison with 2006. This difference represented a reduction in undiagnosed cases of diabetes from 7.1% in 2006 to 3.6% in 2012, and a reduction of impaired fasting glucose (IFG) from 19.0% in 2006 to 8.2% in 2012.

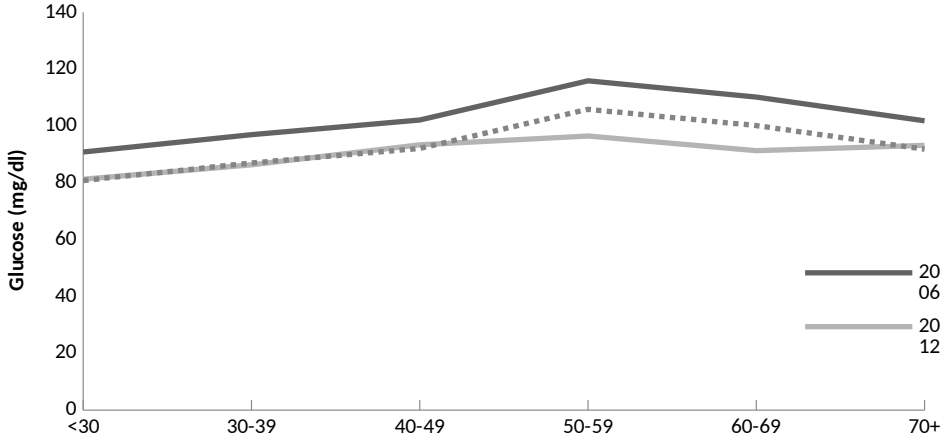
An 11.25 mg/dl reduction was considered too high to be plausible. Still, we explored the possibility that glucose changes could be explained by changes in risk factors between 2006 and 2012. Linear regression models with glucose as dependent variable and survey wave as independent variable were fit, adjusting for age, body mass index, area of residence (urban or rural), and insulin and hypoglycemic drug use. Differences between survey waves remained after adjustment, concluding that differences between 2006 and 2012 were not attributable to differences in main risk factors. An increase in diabetes mortality was also considered a possibility, but it was quickly discarded after comparing the incidence and mortality rates, which in no way could produce a reduction in prevalent cases. After discarding potential causal explanations for the difference between 2006 and 2012 we considered the possibility of measurement bias.

A thorough review of all procedures was conducted to detect critical steps in which plasma glucose quantitation could have been affected. Three steps were considered critical: fasting time prior to drawing blood, invalid quantitation at the laboratory, and degradation of glucose due to glycolysis prior to the separation of plasma. No significant difference in fasting time between survey waves was observed. Available plasma samples from the ENSANUT 2012 were measured again in an independent laboratory, to eliminate the possibility of a laboratory error in measurement; no differences between laboratories were detected. In the absence of differences in these two analyses, the most plausible hypothesis was degradation of samples during field collection. Glucose degrades rapidly in the tube if the intracellular elements are not separated from the blood.(1) While ENSANUT protocols are strict, field conditions are challenging and delays in the time until whole blood tubes were centrifuged could have been prolonged, leading to sample degradation.

Assuming that sample degradation was the mechanism, we further assumed that degradation times have been distributed randomly across ENSANUT participants. To estimate the magnitude of the bias, we identified the subgroup of the adult population less susceptible to glucose changes between 2006 and 2012, that is, adults with no previous diagnosis of diabetes (and no use of insulin/hypoglycemics) and under 50 years of age, as they are less targeted by preventive measures that could lead to glucose reductions. Within this group, we expected to find no significant differences between 2006 and 2012; if observed, the difference would have to be attributable to bias, assuming that the 2006 survey is unbiased and that the population parameter in this group did not change by other reasons between 2006 and 2012.

Figure 1 shows the differences in glucose by age group between 2006 and 2012 in the population with no previous diagnosis of diabetes. In the age-groups under 50 years old we found a stable difference (channel of bias) between 2006 and 2012. After 50 years old, the channel bias gets bigger, which may be associated with preventive measures for adults with diabetes. The difference in glucose between 2006 and 2012 for those under 50 with no previous diagnosis of diabetes was 9.56 mg/dl, and was proposed as a correction factor to be applied to the whole adult population.

**Figure 1: Glucose distribution by age-groups 2006-2012 with no previous diagnosis of diabetes**




To corroborate the impact of the correction against an external source, we used the 2015 Mexico City (CDMX) survey (table 1). To compare both surveys, we selected from ENSANUT adults between 20-69 years old, living in Mexico City. We found that the prevalence from CDMX survey was similar to the ENSANUT prevalence after correcting glucose concentrations, but significantly different to the uncorrected dataset.

*Table 1. Glucose and diabetes prevalence in CDMX survey, ENSANUT 2012 not corrected and 2012 corrected. restricting the sample Mexico City with adults from 20-69 years.*

| | CDMX SURVEY2015 (N=1334) | ENSANUT 2012 not corrected (N=131) | ENSANUT 2012 corrected (N=131) |
|---|---|---|---|
| *Glucose (mg/dl)* | 103.3 (100.7, 105.8) | 94.7 (86.5,102.9) | 104.2 (96.1, 112.4) |
| *Undiagnosed diabetes (%)* | 3.3 (2.5, 4.4) | 2.7 (0.6, 12.0) | 2.7 (0.6, 11.9) |
| *Diagnosed diabetes (%)* | 9.0 (8.8, 9.1) | 9.7 (5.4, 16.7) | 9.7 (5.4, 16.7) |
| *Total diabetes (%)* | 12.2 (11.4, 15.4) | 12.3 (6.2, 23.0) | 12.3 (6.3, 22.9) |
| *Impaired glucose tolerance (%)* | 17.6 (14.9, 20.6) | 7.5 (2.5, 19.8) | 27.7 (17.9, 40.2) |

## References

1. Bruns DE, Knowler WC. Stabilization of Glucose in Blood Samples: Why It Matters. *Clin Chem* 2009;55:850–852.