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COMMENT ON SOFI ET AL.

pH of Drinking Water Influences the Composition of Gut Microbiome and Type 1 Diabetes Incidence. *Diabetes* 2014;63:632–644

Diabetes 2015;64:e19 | DOI: 10.2337/db15-0321

We read with interest the report by Sofi et al. (1) in *Diabetes*. This article is of practical interest to many groups who use the nonobese diabetic (NOD) mouse model. As this group observed, we have also observed that mice that are ordered from The Jackson Laboratory tend to get diabetes sooner than those bred in National Institutes of Health facilities. The results in this article suggest this is partly due to the use of acidic (pH 3.0–3.2) water. If true, it would be a relatively easy way to obtain a more consistent diabetes phenotype for experiments. However, another article was published with data suggesting that acidic water reduces diabetes incidence in NOD mice (2).

Therefore, we decided to repeat the comparison of diabetes incidence with neutral or acidic water in our facility. NOD/ShiLtJt mice were purchased from The Jackson Laboratory and maintained under specific pathogen-free conditions and were fed autoclaved LabDiet JL Rat and Mouse/Auto 6F 5K67 diet, as well as sterile water. Water was acidified by adding hydrochloric acid to achieve a pH range of 3.0–3.3. Breeding pairs were split between neutral and acidified water, and all pups born from these breeding pairs were kept on their specific water. NOD female mice on acidic ($n = 35$) or neutral water ($n = 37$) were followed from 9 weeks old until 30 weeks old for the onset of diabetes, testing urine glucose once a week. A positive test was confirmed by a blood glucose measurement >250 mmol/L. At 30 weeks, 75.7% of NOD mice on neutral water mice became diabetic as compared with

77.1% of NOD mice on acidic water. Significance was calculated using either the Mantel-Cox test (P value = 0.9741) or the Gehan-Breslow-Wilcoxon test (P value = 0.9909). No difference in diabetes timing or incidence was observed. In addition, insulinitis was measured in a separate cohort of mice, and no significant difference in insulinitis score was observed at 9 weeks of age (average score 0.34 for neutral group and 0.26 for low-pH group, P value = 0.2).

In conclusion, although Sofa et al. (1) reported an acceleration of diabetes in NOD mice on acidic water, this result is not universal across different animal facilities. Sofi et al. reported a change in microbiome as the mechanism for the effect of acidic water on diabetes development. One possibility is that the baseline commensals are different at different locations, and this may interact with the acidic water treatment, eliciting different results in different facilities.

Duality of Interest. No potential conflicts of interest relevant to this article were reported.

References

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