Electronic Supplementary Material (ESI) for Analyst.

Alcohol-induced DNA Damage Analysis in *Escherichia coli* by Visualizing Genomic DNA Molecules

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Supplementary Information (SI)

DNA size determination from molecular length: DNA size was determined from stretched DNA lengths. In order to obtain the conversion ratio, we elongated YOYO-1 stained T4 GT7 DNA (165,644 bp, Nippon Gene, Toyama, Japan) on a positively charged surface in a microfluidic channel. The average length was 68.6 ± 9.4 μm from 340 T4 DNA molecules (Fig. S1). Although B-form molecular length for T4 DNA is 56.3 μm from 0.34 nm/bp, YOYO-1 staining increased the contour length approximately 30% and microfluidic stretching condition reduced DNA stretching a little bit depending on fluidic condition.[1] Therefore, we determined the conversion ratio of 0.41 μm/kbp (68.6 μm/ 166 kbp).

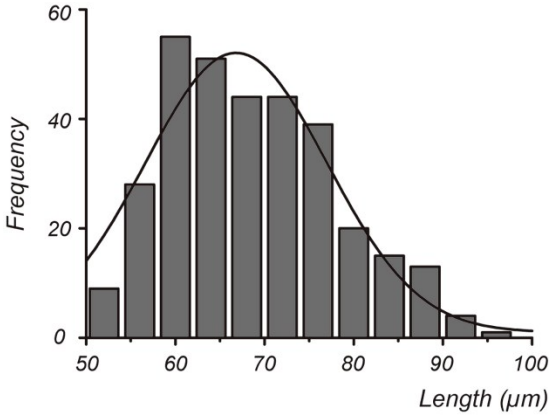

Figure S1. Distribution of measured T4 DNA length (166 kbp).

Pulsed Field Gel Electrophoresis for Damaged *E. coli* Genome: Harvested bacterial cells were washed twice and suspended in 50 mM EDTA (pH 8) to an optical density (OD_{600}) of approximately 3.0. After harvesting bacteria by centrifugation, cell pellets were mixed with 63 μL of 1× TE and 37 μL of 2 % LGT agarose. The mixture was aliquoted by 90 μL in disposable plug mold and solidified in the refrigerator for 30 minutes. Bacteria embedded agarose plugs were incubated in ethanol or alcoholic beverage for 30 minutes. Agarose plug was incubated with proteinase K solution (6 units proteinase K in 300 μL of proteinase K reaction buffer) was added to lyse the bacterial cells for overnight at 50°C. Then, the plugs were washed three times for 1 hour in 1× TE. PFGE was performed using 1 % pulsed field certified agarose gel in 2.2 L 0.5× TBE with yeast chromosome PFG marker. CHEF mammalian genomic DNA plug kit (#170-3591) and 1 % pulsed field certified agarose gel were purchased from Bio-Rad Laboratories (Hercules, CA). The electrophoretic conditions were 2 second initial switch time, 60 second final switch time, 19 hour run time, and 6 V/cm gradient at 14 °C.

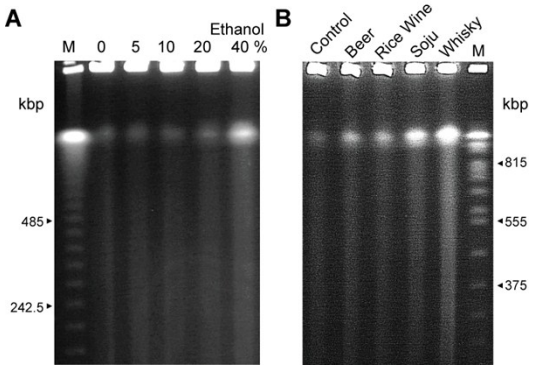

Figure S2. Pulsed field gel electrophoresis of *E. coli* genome after incubation as an agarose plug in ethanol and alcoholic beverage for 30 min. (A) Each lane represents the ethanol percentages and the first lane (M) is lambda ladder PFG marker. (B) Each lane represents four alcoholic beverages: control (0.85 % NaCl solution), beer (5% ethanol), rice wine (13 %

ethanol), soju (20% ethanol), and whiskey (40% ethanol). The last lane (M) represents yeast chromosome marker.

(1) Dimalanta, E. T.; Lim, A.; Runnheim, R.; Lamers, C.; Churas, C.; Forrest, D. K.; de Pablo, J. J.; Graham, M. D.; Coppersmith, S. N.; Goldstein, S.; Schwartz, D. C. *Anal Chem* **2004**, *76*, 5293-5301.