

Differences in diet-microbiome associations dependent on dietary collection methods for proximal and habitual dietary intake

N.M. Simm^{1,2,3}, G.M. Williams^{2,3}, E.C. Hoedt^{1,2,3}, S.J. Caban^{1,2,3}, K.L. Tooley⁴, R. Peterson⁵, K.L. Mudie⁶, G.W. Tyson⁷, P.R. Sternes⁷, S. Keely^{1,2,3}, N.J. Talley^{2,3,8} and K. Duncanson^{2,3,8}

¹School of Biomedical Sciences & Pharmacy, University of Newcastle, Newcastle, New South Wales, Australia

²NHMRC Centre for Research Excellence in Digestive Health, University of Newcastle, Newcastle, New South Wales,

Australia

³Hunter Medical Research Institute, Newcastle, New South Wales, Australia

⁴Defence Science and Technology Group, Department of Defence, Edinburgh, South Australia, Australia

⁵Defence Science and Technology Group, Department of Defence, Melbourne, Victoria, Australia

⁶Defence Science and Technology Group, Department of Defence, Sydney, New South Wales, Australia

⁷Centre for Microbiome Research, School of Biomedical Sciences, Queensland University of Technology, Brisbane,

Queensland, Australia

⁸School of Medicine & Public Health, College of Health, Medicine and Wellbeing, University of Newcastle, New South Wales, Australia

Dietary intake modulates the gut microbiota by providing fermentation substrates. Both microbiota-accessible nutrients and digestible food components have been shown to modulate microbial abundance and function⁽¹⁾. A range of dietary assessment methods are used to investigate diet-microbe interactions, with two commonly used methods being food frequency questionnaires (FFQ) to assess 'habitual' dietary intake and food recalls which measure recent intake proximal to sampling of microbiota. This study aimed to compare dietmicrobiome associations identified from habitual and proximal dietary intake aligned with stool microbiota sampling in a healthy adult cohort. Military trainees (n = 35), and non-military personnel (junior doctors during hospital placement; n = 21) self-reported proximal dietary intake using digital (Easy Diet Diary) or paper-based 24-hr recalls. Habitual intake was assessed using the Comprehensive Nutrition Assessment Questionnaire (CNAQ)⁽²⁾ FFQ. Both measures were assessed at baseline and study completion. Diet recalls matched to the same week of FFQ were analysed using Foodworks $10^{(3)}$. Stool samples were collected for metagenomic shotgun sequencing and annotated against the Microba Life Sciences platform. MaAsLin2 identified linear associations between nutrients and microbe abundance, controlling for total energy intake and individual variation with repeated measures. Thirty dietary variables common to both dietary assessment methods were used in analysis. Mean daily intakes for total energy and macronutrients were not significantly different between habitual and proximal data. Nutrients that differed between methods were polyols (p < 0.001), sugar (p =0.006), sodium (p = 0.03), alcohol (p < 0.001), vitamin A equivalents (p < 0.001), b-carotene equivalents (p < 0.001) and dietary fibre (p = 0.01). Associations between nutrient intake and microbes also differed between dietary collection methods. Most significant associations were found with nutrients measured by 24-hr recall. Mean (M) proximal intake of polyols (M = 0.9 g, standard deviation (SD) = 1.8 g) was significantly associated with increased relative abundance of Akkermansia spp. and CAG460 spp. but not with habitual intake (M =3.4 g, SD = 3.2 g). Proximal alcohol intake (M = 2.5 g, SD = 8.8 g) was associated with CAG1427 spp. and *Collinsella* spp., which was not identified with habitual intake (M = 4.4 g, SD = 6.7 g). In contrast, habitual sugar intake (M = 149 g, SD = 103 g) was associated with Bacteroides spp. and Blautia spp. This association was not evident for proximal intake (M = 112 g, SD = 68 g), suggesting that some dietmicrobiota associations may depend on the dietary assessment method used. These findings demonstrate the relevance of considering both habitual diet and proximal intake when conducting diet-microbiome research. Further analysis will investigate the role of these microbes and further associations between these nutrients and the functional capacity of the microbiota.

References

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