

Supplementary Materials

Profiling the gut and oral microbiota of hormone receptor-positive, HER2-negative metastatic breast cancer patients receiving pembrolizumab and eribulin

Nancy MY Teng¹, Andrea Malfettone², Matthew J Dalby³, Raymond Kiu^{1,3}, David Seki⁴, Tim Robinson⁵, María Gion⁶, Begoña Bermejo⁷, José Manuel Pérez-García^{2,8}, Aleix Prat⁹, Raúl Márquez Vázquez¹⁰, Antonio Llombart-Cussac^{2,11}, Giuseppe Curigliano^{12,13}, Peter Schmid¹⁴, Romualdo Barroso-Sousa¹⁵, Mario Mancino², Eileen Shimizu², Jose Rodríguez-Morató², Leonardo Mina², Lindsay J Hall^{1,3,4,16}, Stephen D Robinson^{1,17}, Javier Cortés^{2,8,18}

¹Gut Microbes&Health, Quadram Institute Bioscience, Norwich NR4 7UQ, UK.

²Medica Scientia Innovation Research (MEDSIR), Barcelona 08005, Spain.

³Institute of Microbiology and Infection, University of Birmingham, Birmingham B12 2TT, UK.

⁴Intestinal Microbiome, School of Life Sciences, ZIEL - Institute for Food&Health Technical University of Munich, Freising D-80333, Germany.

⁵Bristol Medical School, University of Bristol, Bristol BS8 2BN, UK.

⁶Medical Oncology Department, Hospital Universitario Ramón y Cajal, Madrid 28034, Spain.

⁷Medical Oncology, Hospital Clínico de Valencia, INCLIVA, CIBERONC, Medicine Department, Universidad de Valencia, Valencia 46010, Spain.

⁸International Breast Cancer Center (IBCC), Pangaea Oncology, Quirón Group, Barcelona 08017, Spain.

⁹Medical Oncology Department, Hospital Clínic y Provincial de Barcelona, Barcelona 08036, Spain.

¹⁰Servicio de Oncología Médica, MD Anderson Cancer Center, Madrid 28033, Spain.

¹¹Department of medical oncology, Hospital Arnau de Vilanova, FISABIO, Valencia 46800, Spain.

¹²European Institute of Oncology, IRCCS, University of Milano, Milano 20141, Italy.

¹³Department of Oncology and Hemato-Oncology, University of Milano, Milano 20122, Italy.

¹⁴Barts Cancer Institute, Queen Mary University of London, London E1 4NS, UK.

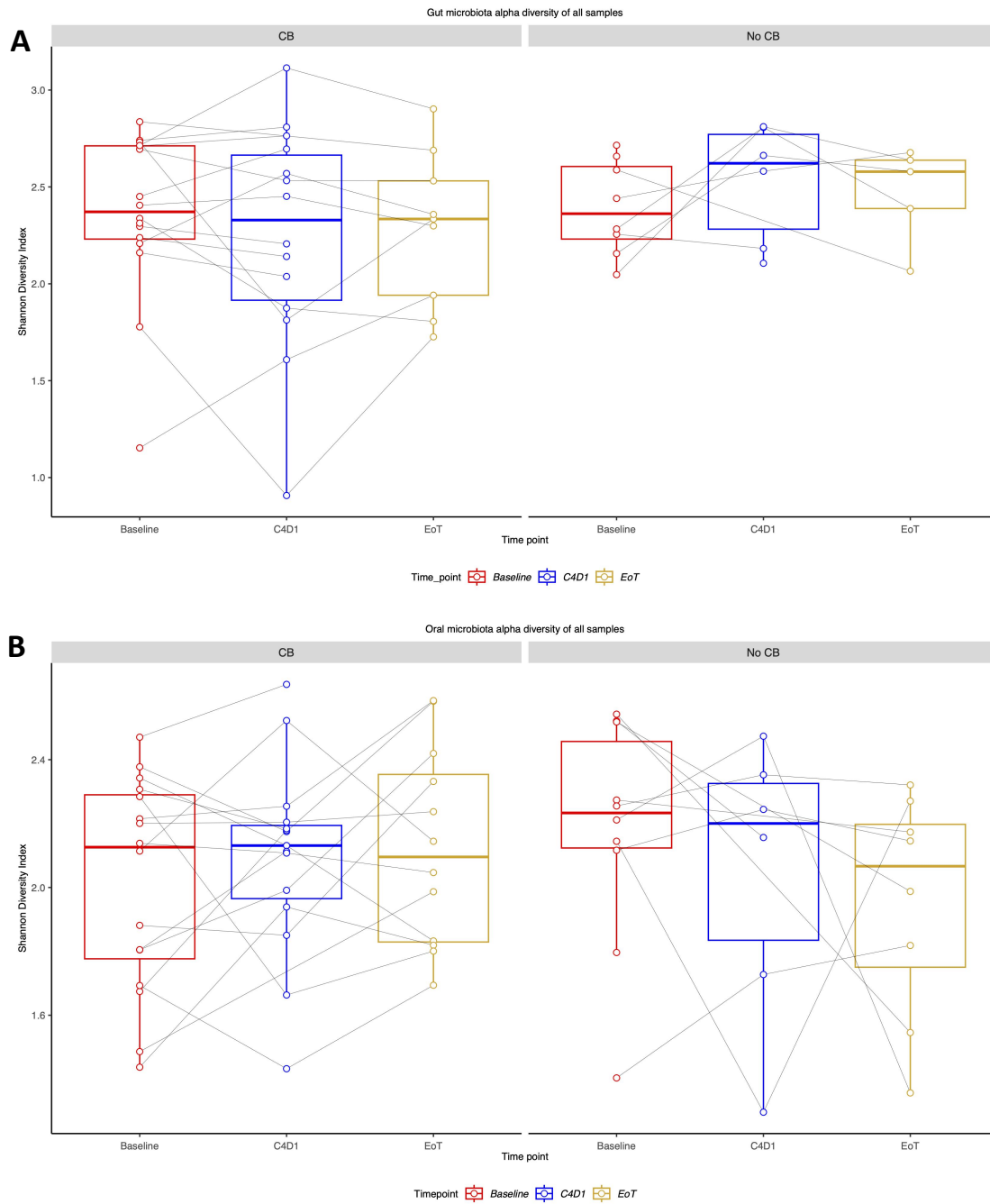
¹⁵Oncology Center, Hospital Sirio-Libanes, Brasília-DF 70200-730, Brazil.

¹⁶Norwich Medical School, University of East Anglia, Norwich NR4 7TJ, UK.

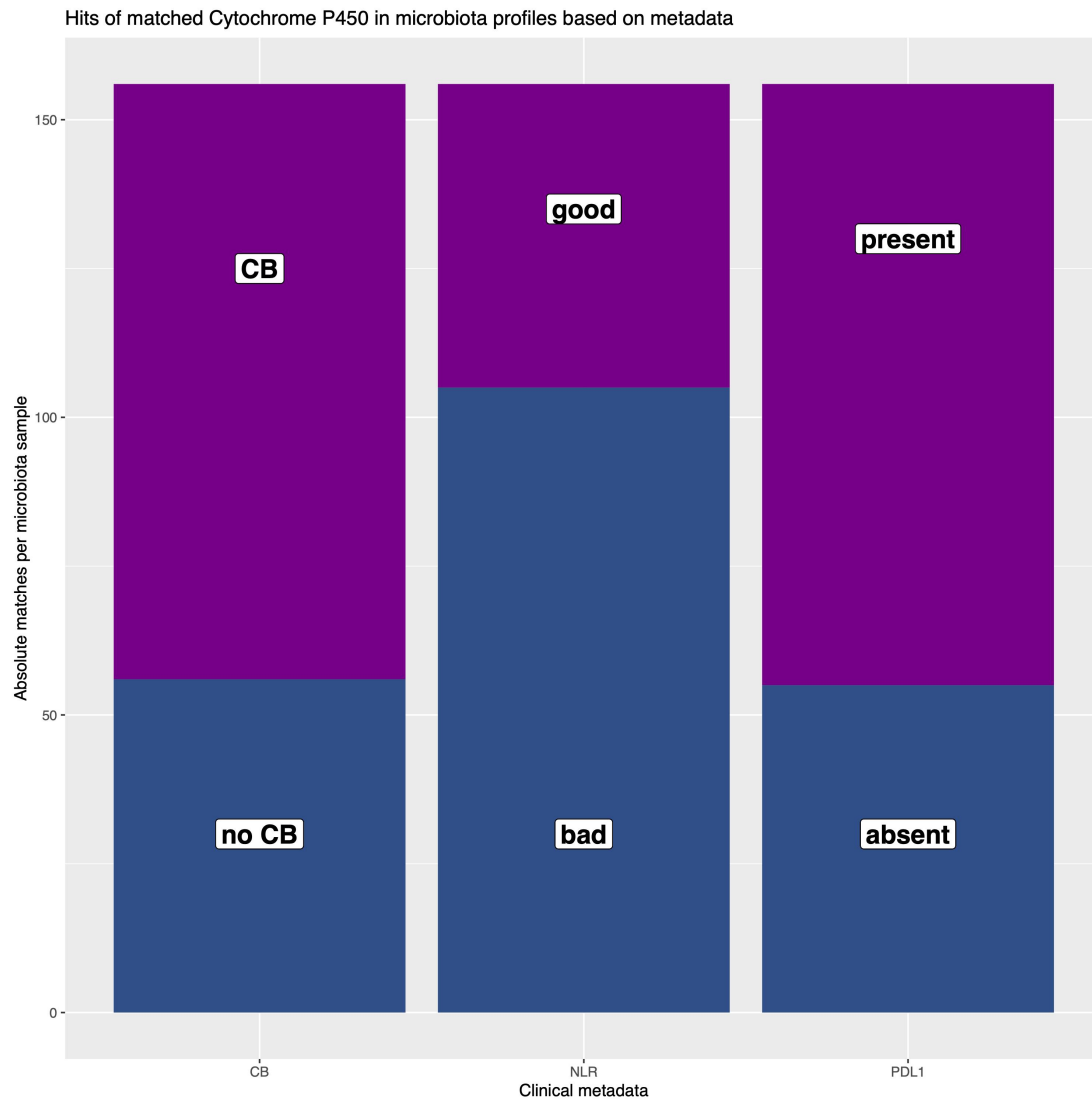
¹⁷School of Biological Sciences, University of East Anglia, Norwich NR4 7TJ, UK.

¹⁸Department of Medicine, Faculty of Biomedical and Health Sciences, Universidad Europea de Madrid, Madrid 28670, Spain.

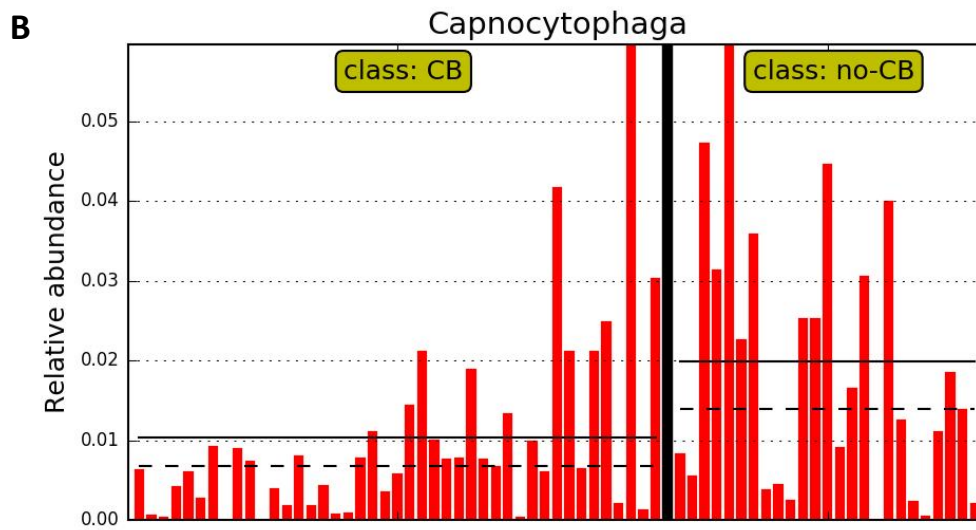
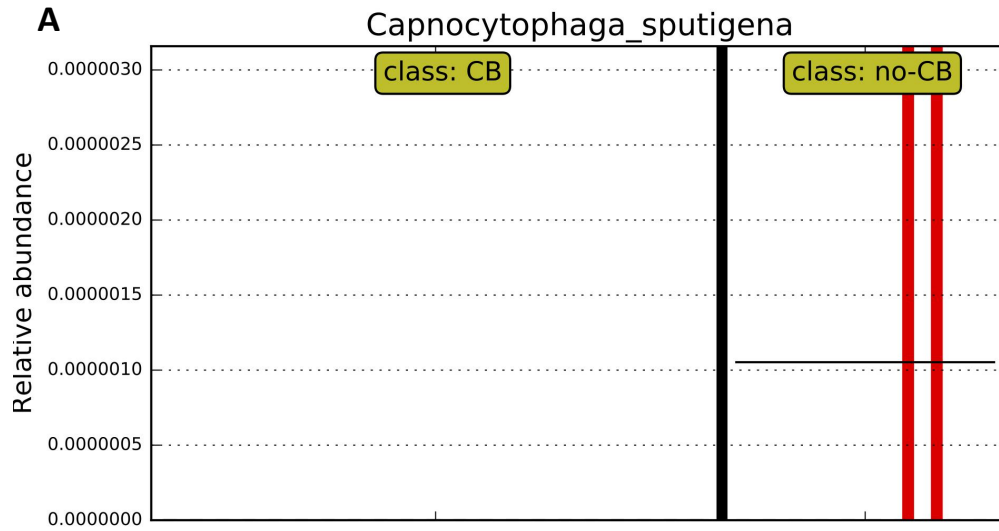
Correspondence to: Dr. Stephen D Robinson, Prof. Lindsay J Hall, Gut Microbes&Health, Quadram Institute Bioscience, Norwich Research Park, Norwich NR4 7UQ, UK. E-mail: stephen.robinson@quadram.ac.uk; lindsay.hall@quadram.ac.uk



Supplementary Figure 1. Study treatment does not confer microbiota toxicity. No significant changes in the gut alpha diversity at the genus level were detected in patients who experienced a clinical benefit (CB) (A). Those patients who did not experience a CB showed a trend toward increasing gut alpha diversity, though not significant. This trend was not observed in the oral alpha diversity, also at the genus level (B). The line connecting the data points denotes samples from the same patient.



Supplementary Figure 2. Number of hits of Cytochrome P450 genes within each CALADRIO metagenome, stratified by clinical parameters: CB, NLR, PD-L1. CB: Clinical benefit; NLR: neutrophil-to-lymphocyte-ratio; PD-L1: programmed cell death ligand 1.



Supplementary Figure 3. Differential feature, Capnocytophaga, determined by LefSe for (A) gut and (B) oral microbiota. Only two fecal samples had a high relative abundance of *Capnocytophaga sputigena*, suggesting it could be a false positive reported in LefSe analysis.

Supplementary Table 1. 57 common genera between the oral and gut microbiota

<i>Acidovorax</i>	<i>Desulfovibrio</i>	<i>Methylobacterium</i>
<i>Acinetobacter</i>	<i>Enterobacter</i>	<i>Mobiluncus</i>
<i>Actinomyces</i>	<i>Enterococcus</i>	<i>Mogibacterium</i>
<i>Aggregatibacter</i>	<i>Faecalibacterium</i>	<i>Mycoplasma</i>
<i>Agrobacterium</i>	<i>Filifactor</i>	<i>Neisseria</i>
<i>Bacillus</i>	<i>Flavobacterium</i>	<i>Odoribacter</i>
<i>Bacteroides</i>	<i>Fusobacterium</i>	<i>Paenibacillus</i>
<i>Bifidobacterium</i>	<i>Gemella</i>	<i>Paracoccus</i>
<i>Blautia</i>	<i>Geobacillus</i>	<i>Porphyromonas</i>
<i>Brevibacillus</i>	<i>Haemophilus</i>	<i>Prevotella</i>
<i>Bulleidia</i>	<i>Klebsiella</i>	<i>Pseudomonas</i>
<i>Butyrivibrio</i>	<i>Lachnospira</i>	<i>Ruminococcus</i>
<i>Campylobacter</i>	<i>Lactobacillus</i>	<i>Shewanella</i>
<i>Capnocytophaga</i>	<i>Lactococcus</i>	<i>Sphingomonas</i>
<i>Chryseobacterium</i>	<i>Lautropia</i>	<i>Staphylococcus</i>
<i>Clostridium</i>	<i>Leptotrichia</i>	<i>Stenotrophomonas</i>
<i>Coprococcus</i>	<i>Leuconostoc</i>	<i>Streptococcus</i>
<i>Desulfobulbus</i>	<i>Macrococcus</i>	<i>Treponema</i>
<i>Desulfomicrobium</i>	<i>Methanobrevibacter</i>	<i>Weissella</i>