

# P3.173: Gut microbiome taxa associated with response to chemo-immunotherapy in NSCLC: a multi-method differential abundance analysis

R. Serna-Blasco<sup>1,2</sup>, A. Rodríguez-Festa<sup>1,2</sup>, A. San Martín<sup>1,2</sup>, E. Nadal<sup>2,3</sup>, J.L. Gonzalez-Larriba<sup>2,4</sup>, A. Martinez-Martí<sup>2,5</sup>, R. Bernabe<sup>2,6</sup>, J. Bosch-Barrera<sup>2,7</sup>, M. Lázaro<sup>2,8</sup>, V. Calvo<sup>1,2</sup>, A. Insa<sup>2,9</sup>, S. Ponce<sup>2,10</sup>, N. Reguart<sup>2,11</sup>, J. De Castro<sup>2,12</sup>, M.R. García Campelo<sup>2,13</sup>, M. Cobo<sup>2,14</sup>, A. Aguilar<sup>2,15</sup>, C. Camps<sup>2,16</sup>, R. López-Castro<sup>2,17</sup>, T. Morán<sup>2,18</sup>, I. Barneto<sup>2,19</sup>, D. Rodríguez - Abreu<sup>2,20</sup>, M. Antoñanzas<sup>2,4</sup>, R. Palmero<sup>2,3</sup>, F. Hernando-Trancho<sup>2,4</sup>, J. Martín-López<sup>1,2</sup>, B. Massutí<sup>2,21</sup>, A. Romero<sup>1,2</sup>, M. Provencio<sup>1,2</sup>

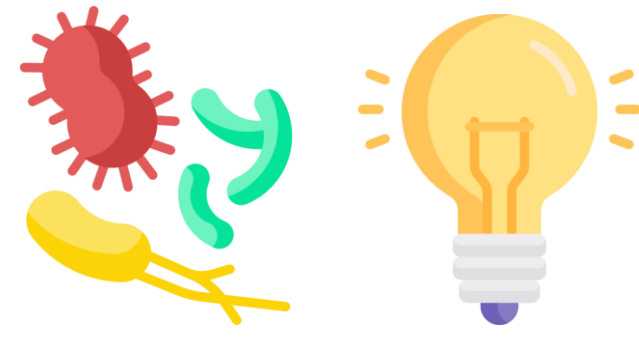
<sup>1</sup>HOSPITAL UNIVERSITARIO PUERTA DE HIERRO MAJADAHONDA, Madrid/ES, <sup>2</sup>Grupo Español de Cáncer de Pulmón (GECp), Barcelona/ES, <sup>3</sup>Institut Català d'Oncologia, L'Hospitalet de Llobregat, Barcelona/ES, <sup>4</sup>HOSPITAL CLÍNICO SAN CARLOS, Madrid/ES, <sup>5</sup>Hospital Universitario Vall d'Hebron, Barcelona/ES, <sup>6</sup>Hospital Universitario Virgen del Rocío, Sevilla/ES, <sup>7</sup>ICO GIRONA - HOSPITAL DR. TRUETA, Girona/ES, <sup>8</sup>Complejo Hospitalario de Vigo, Vigo/ES, <sup>9</sup>HOSPITAL CLÍNICO UNIVERSITARIO DE VALENCIA, Valencia/ES, <sup>10</sup>HOSPITAL UNIVERSITARIO 12 DE OCTUBRE, Madrid/ES, <sup>11</sup>Hospital Clínic de Barcelona, Barcelona/ES, <sup>12</sup>HOSPITAL UNIVERSITARIO LA PAZ, Madrid/ES, <sup>13</sup>Complejo Hospitalario de la Coruña, A Coruña/ES, <sup>14</sup>HOSPITAL REGIONAL UNIVERSITARIO DE MÁLAGA, Málaga/ES, <sup>15</sup>HOSPITAL UNIVERSITARI DEXEUS, Barcelona/ES, <sup>16</sup>HOSPITAL GENERAL UNIVERSITARIO DE VALENCIA, Valencia/ES, <sup>17</sup>HOSPITAL CLÍNICO UNIVERSITARIO DE VALLADOLID, Valladolid/ES, <sup>18</sup>INSTITUT CATALÀ D'ONCOLOGIA - BADALONA, Barcelona/ES, <sup>19</sup>HOSPITAL UNIVERSITARIO REINA SOFIA, Córdoba/ES, <sup>20</sup>HOSPITAL UNIVERSITARIO INSULAR DE GRAN CANARIA, Gran Canaria/ES, <sup>21</sup>HOSPITAL GENERAL UNIVERSITARIO DR. BALMIS DE ALICANTE, Alicante/ES

Check out our  
complementary poster!  
**P3.130**

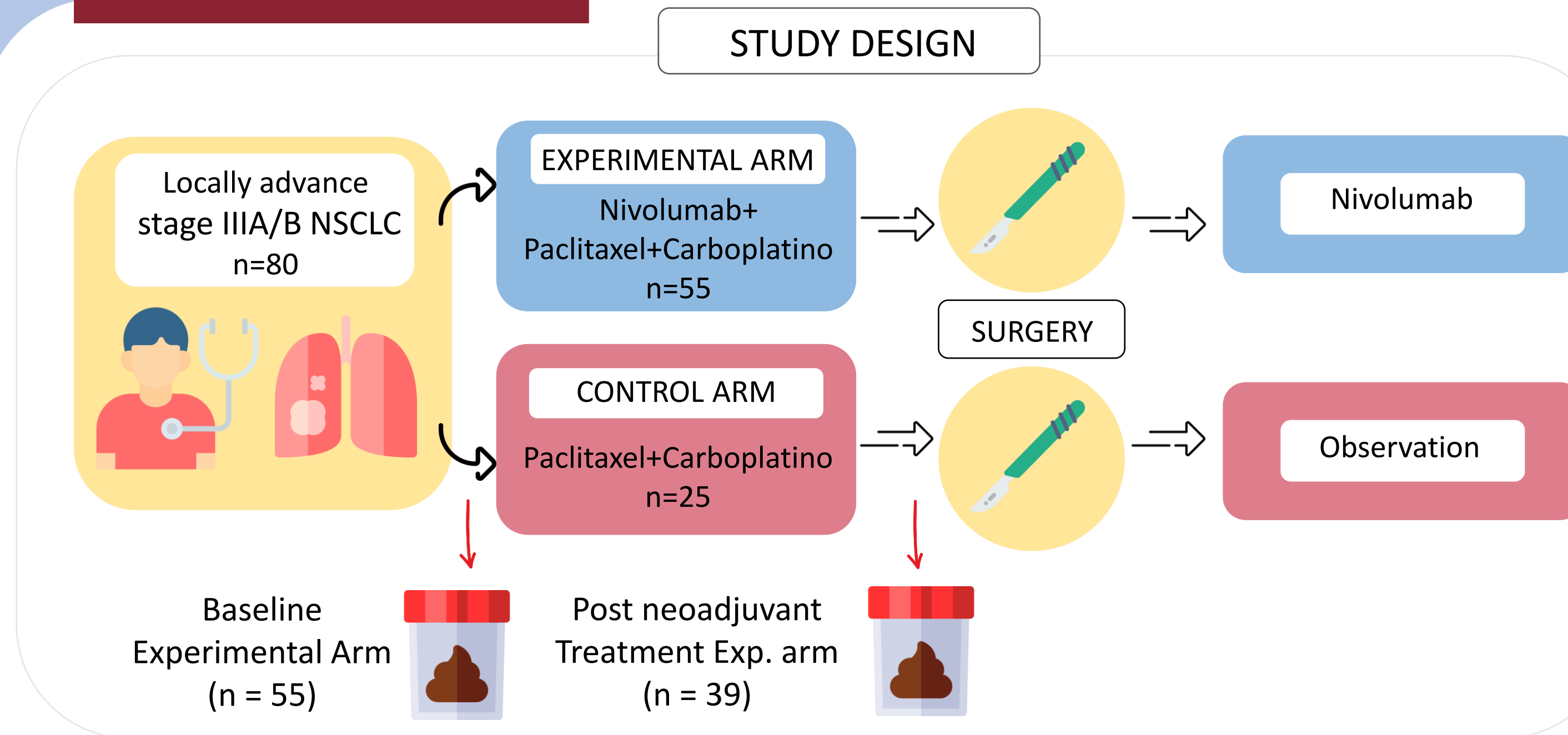
## Introduction

The gut microbiome modulates the immune system and has been associated with clinical outcomes in patients with non-small cell lung cancer (NSCLC) treated with immunotherapy. However, immune-related taxa identified across studies are inconsistent, partly due to the lack of standardized methods for differential abundance analysis (DAA).

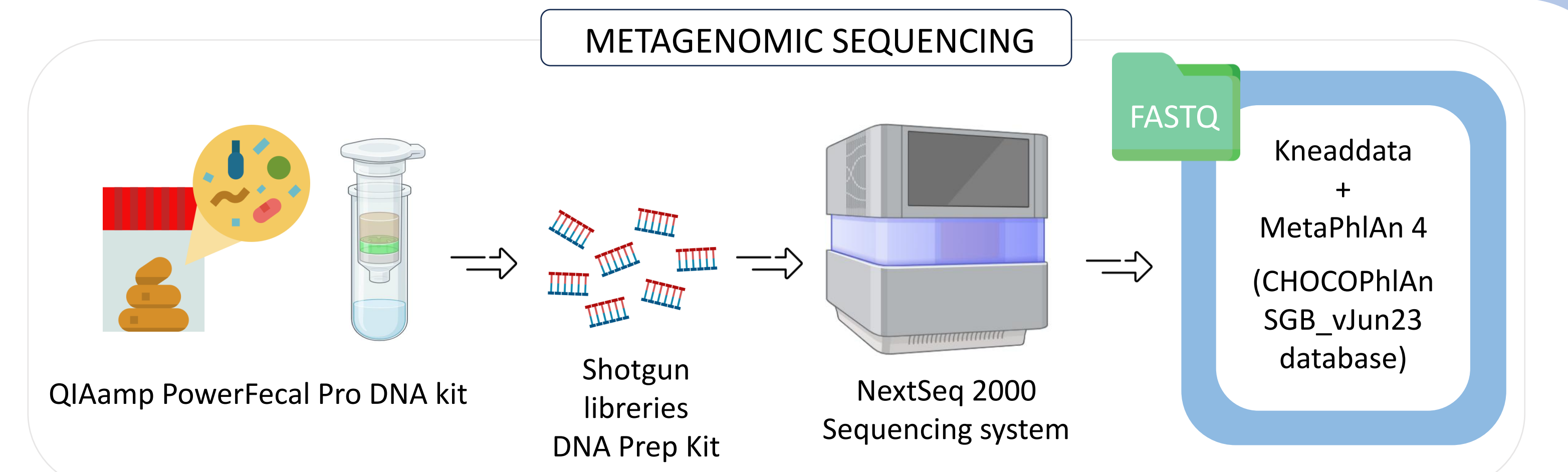
Here, we analyzed the gut microbiome of patients treated with neoadjuvant immunotherapy plus chemotherapy (experimental arm) of the NADIM II trial (NCT03838159), applying multiple statistical approaches based on both relative abundance and estimated counts.



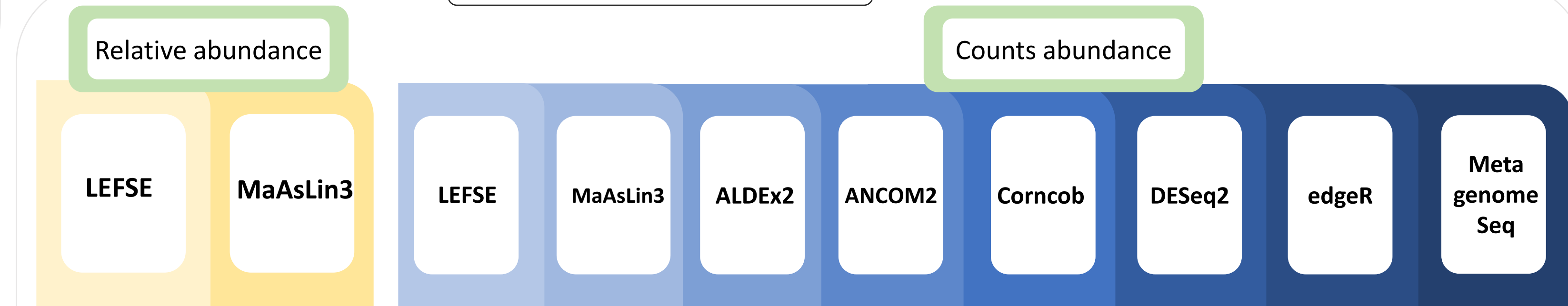
## Methods



Relative abundance data were analyzed using MaAsLin3 and LEfSe for DAA. As many DAA methods rely on count data, estimated absolute counts were also used to perform complementary analyses with ALDEx2, ANCOM2, Corncob, DESeq2, edgeR, LEfSe, MaAsLin3, and metagenomeSeq. Multivariate analyses were conducted with ANCOM2 and MaAsLin3, adjusting for sex, age, BMI, and batch.



## MICROBIOME ANALYSIS

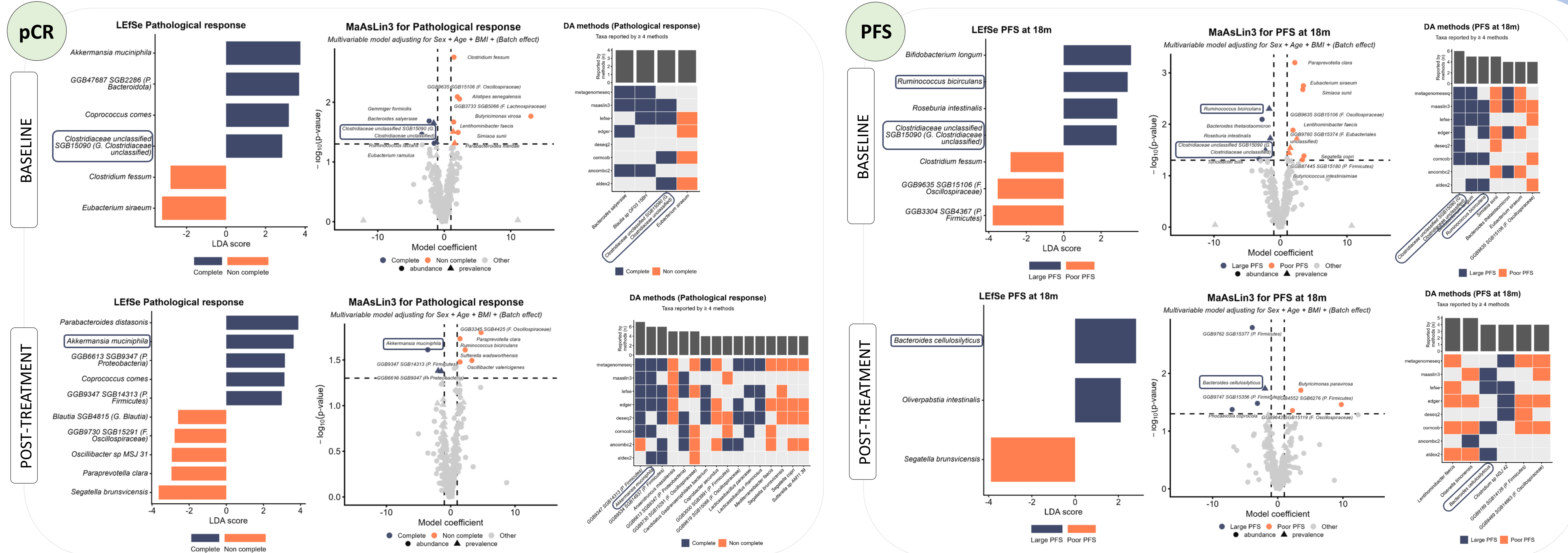


Bacterial taxa were defined as consensus taxa only if identified by both relative abundance-based MaAsLin3 and LEfSe, and by at least four count-based methods.

## Results

According to the consensus taxa definition, an unclassified Clostridiaceae species (SGB15090) was enriched in patients who achieved pCR. Similarly, patients with longer PFS showed enrichment of *Ruminococcus bicirculans* and Clostridiaceae unclassified (SGB15090).

After neoadjuvant treatment, a distinct set of taxa emerged. Notably, *Akkermansia muciniphila* emerged as a significant consensus taxon associated with pCR in post-treatment samples. Moreover, *Bacteroides cellulosilyticus* was associated with patients who achieved an 18-month PFS.



## Conclusion

A consensus-based approach integrating multiple statistical methods identified robust taxa associated with pCR and survival outcomes. These findings highlight the importance of methodological standardization and support the use of integrative analytical strategies to improve the reproducibility of microbiome-based biomarkers.



Estudio financiado por Bristol-Myers Squibb y GECp. Cofinanciado por el Instituto de Salud Carlos III, con código PI24/01725 - PI25/01704 (Resolución de la Dirección del Instituto de Salud-Carlos III, O.A., M.P. de 27 de diciembre de 2024 - 5 de diciembre de 2025) por la que se conceden subvenciones para Proyectos de I+D+i en salud, y Cofinanciado por la Unión Europea.

laboratorio.biopsia.liquida@idihim.org  
arodriguez@idiphim.org  
0000-0003-3344-652X