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# A faecal microbiota signature with high specificity for pancreatic cancer

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## ABSTRACT

**Background** Recent evidence suggests a role for the microbiome in pancreatic ductal adenocarcinoma (PDAC) aetiology and progression.

**Objective** To explore the faecal and salivary microbiota as potential diagnostic biomarkers.

**Methods** We applied shotgun metagenomic and 16S rRNA amplicon sequencing to samples from a Spanish case–control study (n=136), including 57 cases, 50 controls, and 29 patients with chronic pancreatitis in the discovery phase, and from a German case–control study (n=76), in the validation phase.

**Results** Faecal metagenomic classifiers performed much better than saliva-based classifiers and identified patients with PDAC with an accuracy of up to 0.84 area under the receiver operating characteristic curve (AUROC) based on a set of 27 microbial species, with consistent accuracy across early and late disease stages. Performance further improved to up to 0.94 AUROC when we combined our microbiome-based predictions with serum levels of carbohydrate antigen (CA) 19–9, the only current non-invasive, Food and Drug Administration approved, low specificity PDAC diagnostic biomarker. Furthermore, a microbiota-based classification model confined to PDAC-enriched species was highly disease-specific when validated against 25 publicly available metagenomic study populations for various health conditions (n=5792). Both microbiome-based models had a high prediction accuracy on a German validation population (n=76). Several faecal PDAC marker species were detectable in pancreatic tumour and non-tumour tissue using 16S rRNA sequencing and fluorescence *in situ* hybridisation.

**Conclusion** Taken together, our results indicate that non-invasive, robust and specific faecal microbiota-based screening for the early detection of PDAC is feasible.

## Significance of this study

### What is already known about this subject?

- Pancreatic ductal adenocarcinoma (PDAC) is on the rise worldwide, posing a high disease burden and mortality rate, yet accurate, non-invasive diagnostic options remain unavailable.
- Alterations in the oral, faecal and pancreatic microbiome composition have been associated with an increased risk of PDAC.

### What are the new findings?

- Stool microbiota-based classifiers are described that predict PDAC with high accuracy and specificity, independent of disease stage, with potential as agents for non-invasive diagnostics.
- A faecal metagenomic classifier identified PDAC with an accuracy of 0.84 area under the receiver operating characteristic curve (AUROC) in a Spanish cohort, based on 27 species. The accuracy improved to up to 0.94 AUROC when combined with the less specific carbohydrate antigen (CA) 19–9 serum marker.
- The classifier was validated in an independent German PDAC cohort (0.83 AUROC), and PDAC disease specificity was confirmed against 25 publicly available metagenomic study populations with various health conditions (n=5792).
- The presence of marker taxa enriched in faecal samples (*Veillonella*, *Streptococcus*, *Akkermansia*) and also taxa with differential abundance in healthy and tumour pancreatic tissues (*Bacteroides*, *Lactobacillus*, *Bifidobacterium*) was validated by fluorescence *in situ* hybridisation.

## Significance of this study

**How might it impact on clinical practice in the foreseeable future?**

- ▶ Faecal microbiome-based detection of PDAC may provide a non-invasive, cost-effective and robust approach to early PDAC diagnosis.
- ▶ The presented PDAC-specific microbiome signatures, including links between microbial populations across tissues, provide novel microbiome-related hypotheses regarding disease aetiology, prevention and possible therapeutic intervention.

**INTRODUCTION**

Pancreatic ductal adenocarcinoma (PDAC) is the most common form of pancreatic cancer and a major cause of cancer-related deaths despite relatively low incidence rates.<sup>1,2</sup> The high lethality of PDAC is a consequence of both late diagnosis and limited therapeutic options<sup>3</sup>: symptoms are unspecific and often emerge only during late disease stages, at which point tumours can be either locally non-resectable or present as metastatic disease. At present, PDAC is diagnosed using imaging tests.<sup>4</sup> Sensitive and affordable tests for an early detection of PDAC could therefore improve outcome. PDAC markers have been explored in pancreatic tissue,<sup>5</sup> urine<sup>6,7</sup> and serum.<sup>8,9</sup> Yet to date, the sole Food and Drug Administration (FDA)-approved PDAC biomarker remains serum carbohydrate antigen (CA) 19-9. CA19-9 has limited disease specificity as levels can be elevated in several other concomitant conditions (eg, biliary obstruction) and is therefore mostly used as a marker for PDAC surveillance, rather than screening or diagnosis.<sup>10–14</sup>

PDAC has a complex aetiology, with established risk factors that include age, chronic pancreatitis, diabetes mellitus, obesity, asthma, blood group and lifestyle (eg, smoking and heavy alcohol consumption).<sup>15,16</sup> The role of these risk factors in PDAC aetiology may also be complemented—or sometimes indeed mediated—by alterations in the microbiome. For example, poor oral hygiene and periodontitis have been associated with an increased PDAC risk,<sup>17</sup> an observation that also extends to periodontitis- and caries-associated microbial species.<sup>18,19</sup> Shifts in these species are sometimes part of wider compositional changes in the oral microbiome<sup>20,21</sup> or have been explored as PDAC risk factors in their own right.<sup>22</sup> Similarly, microbial composition in the gut<sup>23–25</sup> and duodenum,<sup>26,27</sup> quantified via 16S rRNA amplicon sequencing, have previously been linked to PDAC risk.

The human pancreas harbours a microbiome that shares species with the mouth and the gut,<sup>25,28–32</sup> although its exact composition has remained elusive owing to the challenges associated with contamination control in low bacterial biomass samples.<sup>33</sup> In murine models, microbes originating from the intestine can contribute to carcinogenesis in the pancreatic duct,<sup>25,30</sup> suggesting a role for the microbiome in PDAC aetiology and progression that was recently extended to fungi.<sup>34</sup> Moreover, the pancreatic tumour microbiome may also be associated with disease progression and long-term survival in patients with PDAC.<sup>31</sup>

However, the translation of these advances into PDAC-specific microbiome signatures for clinical applications has so far remained largely unexplored. Here, we present the identification of robust, specific microbial PDAC signatures based on a metagenomic survey of a Spanish (ES) study population of 57 newly diagnosed and treatment-naïve patients

with PDAC, 29 patients with chronic pancreatitis (CP), and 50 matched controls. We sampled saliva, faeces, pancreatic normal and tumour tissue and assessed microbial composition using whole-genome shotgun metagenomics, 16S rRNA amplicon sequencing, and fluorescence *in situ* hybridisation (FISH) assays. The best discrimination between patients with PDAC and non-PDAC subjects was achieved by statistical models based on a set of 27 faecal microbial species that could be quantified in a targeted manner in a diagnostic setting. The prediction accuracy of microbiome-based models was confirmed in an independent German (DE) PDAC validation population including 44 patients with PDAC and 32 controls and was further improved when combined with serum levels of CA19-9. We further validated the disease specificity of these models against existing data from 25 studies (n=5792) of nine diseases.<sup>35–59</sup> Several of the PDAC-enriched species were also detected in cancer tissue, with possible links to oral and intestinal populations, supporting their potential role in PDAC pathogenesis, as previously reported.<sup>25,30,31,34</sup>

**METHODS****Subject recruitment and sample collection**

A case-control design was applied. Subjects were prospectively recruited between 2016 and 2019 from the Hospital Ramón y Cajal in Madrid and Hospital Vall d'Hebron in Barcelona, Spain, using the same protocols for biological sample collection, processing and storage. Subjects with newly diagnosed PDAC (n=57), aged >18 years, were identified prior to any cancer treatment. Subjects in whom PDAC was suspected were recruited, and sampling was done before any treatment. Patients with chronic pancreatitis (CP, n=29) were recruited from the same hospitals. Controls matched for age, gender and hospital were selected from inpatients with a primary diagnosis for hospital admission not related to PDAC risk factors. Participants incapable of participating in the study owing to impairment of physical ability were excluded. Institutional review board ethical approval (CEI PI 26 2015-v7) and written informed consent were obtained from participating centres and study participants, respectively. Epidemiological and lifestyle data were collected by trained monitors during face-to-face interviews through a structured questionnaire. Clinical data, including stage of the diseases and follow-up data, were retrieved from hospital charts by the same monitors, likewise using structured questionnaires. Recorded jaundice status was additionally confirmed and extended by direct bilirubin measurements from blood samples in CNIO, Madrid. All data were entered, edited and managed using REDCap. Missing lifestyle and medication values in the metadata (missing overall in 3.1%) were imputed using a random forest-based algorithm for missing data imputation called missForest (n=100 trees).<sup>60</sup> The imputation accuracy was high according to the imputation error estimate (mean out-of-bag error=0.12). Serum CA19-9 levels were analysed by electrochemiluminescence immunoassay (ECLIA, Roche Diagnostics, Germany) following the manufacturer's instructions in the Institute of Laboratory Medicine and Pathobiochemistry, Marburg, Germany. Each sample was assayed in duplicate, with positive controls assayed in each plate (online supplemental table S1).

Stool and saliva (mouthwash) samples were preserved in RNALater and stored at 4°C immediately for 12 hours, then transferred to –20°C for another 24 hours, and then stored at –80°C until DNA extraction. Tumour and non-affected tissue samples were collected during surgery for a subset of individuals, immediately flash-frozen in liquid nitrogen after pathological

assessment, and preserved at  $-80^{\circ}\text{C}$ . All the samples were shipped on dry ice.

An independent validation population was recruited at the Department of Surgery, University Hospital of Erlangen (32 PDAC and 32 control samples) and Section for Translational Hepatology, Department of Internal Medicine I, Goethe University Clinic, Frankfurt (12 PDAC samples) using the same protocols for biological sample collection, processing and storage. Matched controls were selected from inpatients with a primary diagnosis for hospital admission not related to PDAC risk factors. The study was approved by the local ethics committees (SGI-3-2019, 451\_18 B), and written informed consent from study participants was obtained. Clinical data, including disease stage and follow-up data, were retrieved from the clinical records of the hospital charts of the respective patients (online supplemental table S2). Serum CA19-9 levels were analysed by a routine immunoassay (Roche Diagnostics, Germany) following the manufacturer's instructions. Stool samples were preserved in OMNIgene-Gut OM-200 vials (Steinbrenner Laborsysteme GmbH, Germany) and stored at  $-80^{\circ}\text{C}$  immediately until DNA extraction.

### Sample processing

Faecal and salivary samples were thawed on ice, aliquoted, and genomic DNA was extracted using the Qiagen Allprep PowerFecal DNA/RNA kit according to the manufacturer's instructions (Qiagen, Hilden, Germany). Genomic DNA from pancreatic tumorous and non-tumorous tissue samples was extracted using the Qiagen DNeasy blood and tissue kit in a protocol modified from Del Castillo *et al.*<sup>26</sup>: cells were lysed mechanically (with 5 mm stainless steel beads at 25 Hz for 150 s), followed by lysozyme treatment (20 mg/mL) and protease and RNase digestion ( $56^{\circ}\text{C}$  for 2 h). All samples were randomly assigned to extraction batches. To account for potential bacterial contamination of extraction, polymerase chain reaction (PCR) and sequencing kits, we included negative controls (extraction blanks) with each tissue DNA extraction batch (online supplemental figure 1).

### 16S rRNA amplicon sequencing

Pancreatic tissue DNA was enriched for 16S rRNA in a preamplification PCR using primers 331F ( $5^{\prime}\text{-TCCTACGGGAGGCAG-CAGT-3}^{\prime}$ )<sup>61</sup> and 979R ( $5^{\prime}\text{-GGTCTKCGCGTTGCWTC-3}^{\prime}$ )<sup>62</sup>. The cycling conditions consisted of an initial template denaturation at  $98^{\circ}\text{C}$  for 2 min, followed by 30 cycles of denaturation at  $98^{\circ}\text{C}$  for 10 s, annealing at  $65^{\circ}\text{C}$  for 20 s, extension at  $72^{\circ}\text{C}$  for 30 s and a final extension at  $72^{\circ}\text{C}$  for 10 min. This was followed by a size-selective cleanup using SPRIselect magnetic beads (0.8 left-sized; Beckman Coulter, Brea, California, USA). Faecal and salivary DNA were not preamplified.

Targeted amplification of the 16S rRNA V4 region (primer sequences F515  $5^{\prime}\text{-GTGCCAGCMGCCGCGGTAA-3}^{\prime}$  and R806  $5^{\prime}\text{-GGACTACHVGGGTWTCTAAT-3}^{\prime}$ )<sup>63</sup> was performed using the KAPA HiFi HotStart PCR mix (Roche, Basel, Switzerland) in a two-step barcoded PCR protocol (NEXTflex 16S V4 Amplicon-Seq Kit; Bioo Scientific, Austin, Texas, USA) with minor modifications from the manufacturer's instructions. PCR products were pooled, purified using size-selective SPRIselect magnetic beads (0.8 left-sized) and then sequenced at  $2\times 250$  bp on an Illumina MiSeq (Illumina, San Diego, California, USA) at the Genomics Core Facility, European Molecular Biology Laboratory, Heidelberg.

### 16S rRNA amplicon data processing

Raw reads were quality trimmed, denoised and filtered against chimeric PCR artefacts using DADA2.<sup>64</sup> The resulting exact amplicon sequence variants (ASVs) were taxonomically classified and mapped to a reference set of operational taxonomic units (OTUs) at 98% sequence similarity using MAPseq.<sup>65</sup> Reads that did not confidently map to the reference were aligned to bacterial and archaeal secondary structure-aware small subunit rRNA models using Infernal<sup>66</sup> and clustered into OTUs with 98% average linkage using HPC-CLUST,<sup>67</sup> as described previously.<sup>68</sup> As a result, we obtained taxa tables at two resolutions: 100% identical ASVs and 98% open-reference OTUs; unless otherwise indicated, analyses in the main text refer to OTUs.

Count tables were noise filtered by removing samples retaining less than 500 reads and taxa observed in fewer than five samples; this removed 2.5% of total reads from the dataset. For 18 salivary samples, technical replicates were merged after confirming that they strongly correlated with community composition. For pancreatic tissue and tumour samples, ASVs observed in negative control samples were removed, as were reads mapping to known reagent kit contaminants.<sup>33</sup> After these steps, we retained 308 16S rRNA amplicon samples from 143 subjects for further analyses (130 salivary, 118 faecal, 20 of unaffected pancreatic tissue, 23 of tumour tissue with 17 matching PDAC tissue samples).

### Shotgun metagenomic sequencing

Metagenomic libraries for 212 faecal and 100 salivary samples were prepared using the NEB Ultra II and SPRI HD kits, depending on the concentration of starting material, with a targeted insert size of 350, and sequenced on an Illumina HiSeq 4000 platform (Illumina, San Diego, California, USA) in  $2\times 150$  bp paired-end setup to a target depth of 8 Gbp per sample at the Genomics Core Facility, European Molecular Biology Laboratory, Heidelberg. Sequencing statistics for each sample are provided in the associated git repository (<https://github.com/psecekartal/PDAC.git>). For three salivary and one faecal samples, technical replicates were merged after confirming that they strongly correlated in community composition.

### Metagenome data processing

Metagenomic data were processed using established workflows in NGLess v0.7.1.<sup>69</sup> Raw reads were quality trimmed ( $\geq 45$  bp at Phred score  $\geq 25$ ) and filtered against the human genome (version hg19, mapping at  $\geq 90\%$  identity across  $\geq 45$  bp). The resulting filtered reads were mapped ( $\geq 97\%$  identity across  $\geq 45$  bp) against the representative genomes of 5306 species-level genome clusters obtained from the proGenomes database v2.<sup>70</sup>

Taxonomic profiles were obtained using the mOTU profiler v2.5<sup>71</sup> and filtered to retain only species observed at a relative abundance  $\geq 10^{-5}$  in  $\geq 2\%$  of samples. Gene functional profiles were obtained from mappings against a global microbial gene catalogue (GMGCv1, Coelho *et al.*<sup>72</sup>, <http://gmgc.embl.de/>), by summarising read counts from eggNOG v4.5<sup>73</sup> annotations to orthologous groups and KEGG modules. Features with a relative abundance of  $\geq 10^{-5}$  in  $\geq 15\%$  of samples were retained for further analyses.

### Microbiome data statistical analyses

All data analyses were conducted in the R Statistical Computing framework v3.4 or higher.

Rarefied per-sample taxa diversity ('alpha diversity', averaged over 100 rarefaction iterations) was calculated as the effective number of taxa with Hill coefficients of  $q=0$  (ie, taxa richness),

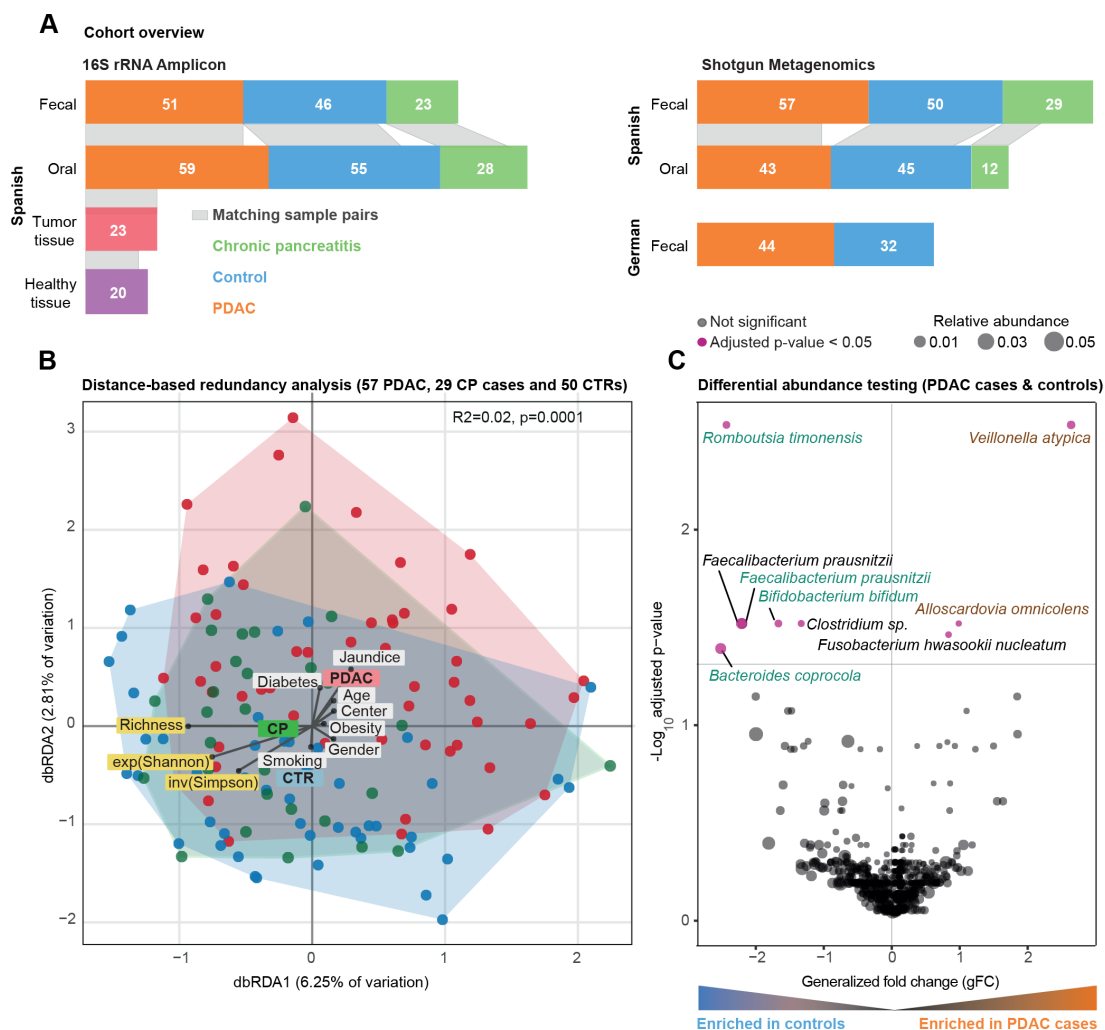
$q=1$  (exponential of Shannon entropy) and  $q=2$  (inverse Simpson index), and evenness measures as ratios thereof. Unless otherwise stated, results in the main text refer to taxa richness. Differences in alpha diversity were tested using analysis of variance (ANOVA) followed by post hoc tests and Benjamini-Hochberg correction, as specified in the main text.

Between-sample differences in community composition ('beta diversity') were quantified as Bray-Curtis dissimilarity on raw or square-root transformed counts, abundance-weighted Jaccard index, and abundance-weighted and unweighted TINA index, as described previously.<sup>74</sup> Trends between these indices were generally consistent, unless otherwise stated. Results are reported for Bray-Curtis dissimilarities on non-transformed data. Associations of community composition to microbiome-external factors were quantified using the 'adonis2' implementation of PERMANOVA and distance-based redundancy analysis in the R package *vegan* v2.5.<sup>75</sup> To quantify potentially confounding univariate links between the abundance of individual taxa and subject-specific

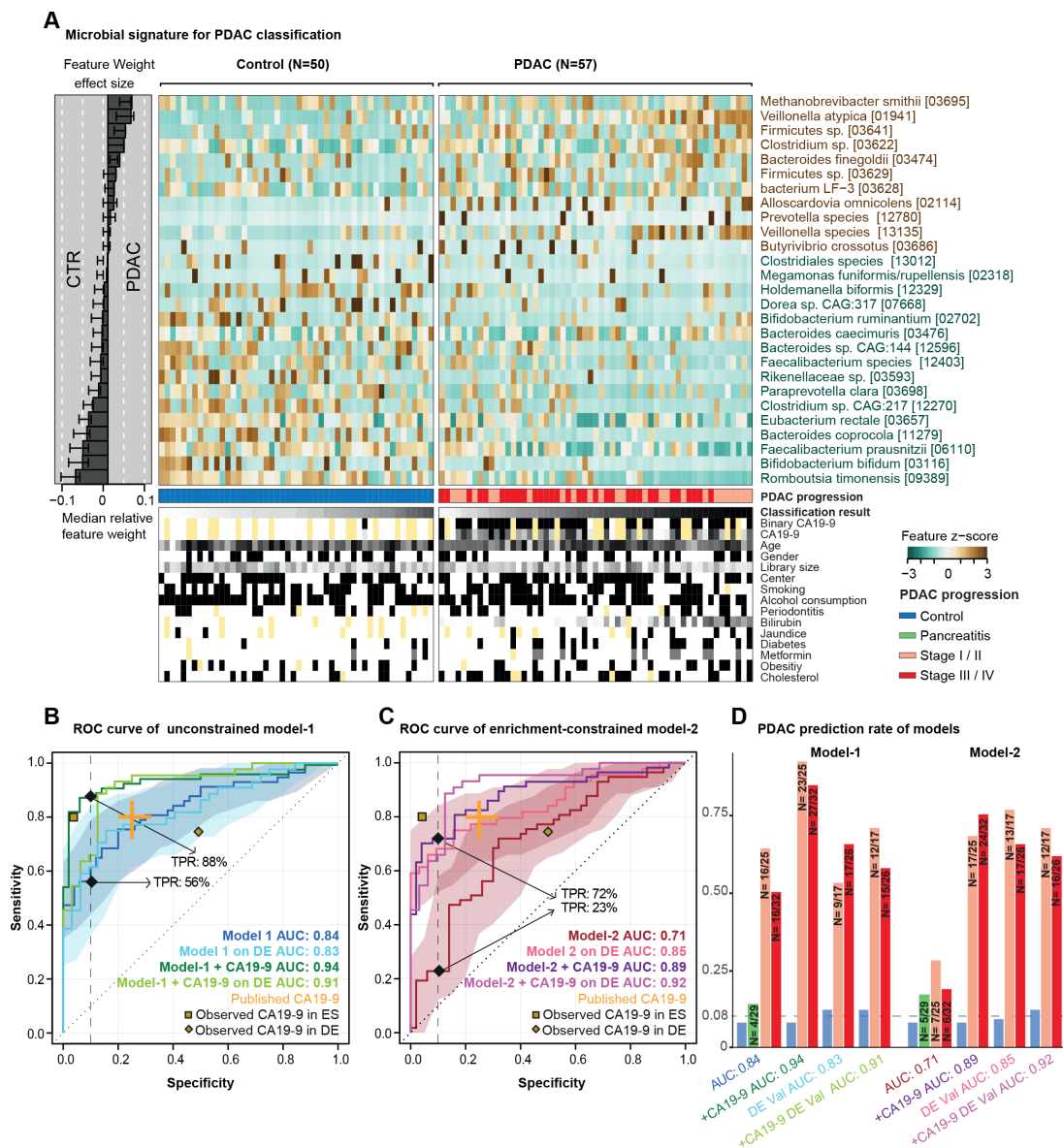
variables (see main text), we performed either ANOVA or non-parametric Kruskal-Wallis tests, depending on abundance distributions (online supplemental figure 2-3 and online supplemental table S4-S5). Bilirubin levels were measured from blood samples, and jaundice status was confirmed by clinical records. Owing to missing jaundice status for several individuals, values used for further analysis were imputed from existing data (figure 1, online supplemental table S1-S3).

### Multivariable statistical modelling and model evaluation

In order to train multivariable statistical models for the prediction of pancreatic cancer, we first removed taxa with low overall abundance and prevalence (abundance cut-off point: 0.001). Then, features were normalised by log<sub>10</sub> transformation (to avoid infinite values from the logarithm, a pseudo-count of 1e-05 was added to all values) followed by standardisation as centred log-ratio (log<sub>10</sub>clr). Data were randomly split into test and



**Figure 1** Community analysis of Spanish faecal microbiome data. (A) Study population overview. Grey bands between the bar plots indicate samples of matching body sites within individuals. (B) Bray-Curtis distance-based redundancy analysis (dbRDA) of pancreatic ductal adenocarcinoma (PDAC), chronic pancreatitis (CP) and control (CTR) faecal microbiome data in a Spanish (ES) cohort. PDAC samples are shown as red coloured circles, patients with CP as green and controls as blue. Richness, exponential Shannon (exp(Shannon)) and inverse Simpson (inv(Simpson)) diversity measures are also visualised with arrows similarly to tested metadata variables. The distance of the meta-variable from the centre represents the confounding effect size (see 'Methods'). (C) Wilcoxon test results of ES faecal microbiome data to test enriched taxa between PDAC and control cases (see 'Methods'). Y-axis is log<sub>10</sub>(FDR corrected p values), X-axis is generalised fold change, and dot size represents the relative abundance of a given species. Red dots represent significantly differentially abundant species in either group, while black dots show non-significant species after FDR correction. Green and brown-coloured species are selected in metagenomic model-1 as predictors of PDAC. FDR, false discovery rate.



**Figure 2** Predictive microbiome signatures of pancreatic ductal adenocarcinoma (PDAC). (A) Normalised abundance of 27 selected species in the faecal microbiome across samples shown as a heat map. The right panel represents the contribution of each selected feature to the overall model-1, and the robustness (the percentage of models in which the feature is included as predictor) of each feature is presented as percentage. Classification scores from cross-validation of each individual and condition for tested meta-variables are displayed at the bottom of the panel, yellow representing missing information. (B–D) Internal cross-validation results of unconstrained model-1 (without feature selection), enrichment-constrained model-2 (constrained to positive features) and combination of carbohydrate antigen (CA)19-9 (using a threshold of 37  $\mu\text{L}/\text{mL}$ ) with microbial features (see ‘Methods’) are shown as receiver operating characteristic (ROC) curve with 95% CI shaded in corresponding colour. True positive rates (TPRs) are given as a percentage at a 90% specificity cut-off. Validation of all models on an independent German (DE) PDAC test population ( $n=76$ ) is represented as well. Published CA19-9 accuracy from a meta-study shown in orange. The yellow dots represent observed CA19-9 accuracies in our populations (data available for 33/50 controls (CTRs) and 44/57 patients with PDAC in the Spanish (ES) and for 8/32 CTRs and 44/44 patients with PDAC in the German (DE) population) (D) TPRs of all models at different PDAC progression stages and in addition, the false-positive rate for patients with chronic pancreatitis and controls at a 90% specificity cut-off are shown as bar plots. Stages I and II and stages III and IV are combined owing to the overall low sample size. The number of predicted cases compared with the total is also shown on the top of each bar. DE-Val, German validation population.

training sets in a 10 times repeated 10-fold cross-validation. For each test fold, the remaining folds were used as training data to train an L1-regularised (LASSO) logistic regression model<sup>76</sup> using the implementation within the Liblinear R package v2.10.<sup>77</sup> The trained model was then used to predict the left-out test set and finally, all predictions were used to calculate the area under the receiver operating characteristics curve (AUROC) (figure 2).

In a second approach, features were filtered within the cross-validation (that is, for each training set) by first calculating the single-feature AUROC and then removing features with an AUROC  $< 0.5$ , thereby selecting features enriched in PDAC (‘enrichment-constrained’ model).

In order to combine the predictions from the microbiome-based machine learning models with the CA19-9 marker,

the coded CA19-9 marker (1 for positive, 0 for negative or not available) was added to the mean predictions from the repeated cross-validation runs, resulting in an OR combination. Alternatively, the AND combination was calculated by multiplying the predictions with the CA19-9 marker. ROC curves and AUROC values were calculated for both combinations using the pROC R package v1.15.<sup>78</sup> The 95% CI is shaded in corresponding colour and specified in figure legends for each ROC curve.

The trained ES metagenomic classifiers for PDAC were then applied to the DE dataset after applying a data normalisation routine, which selects the same set of features and uses the same normalisation parameters (for example, the mean of a feature for standardisation by using the frozen normalisation functionality in SIAMCAT) as in the normalisation procedure from the ES pancreatic cancer dataset. For this analysis, the cut-off point for the predictions was set to a false-positive rate of 10% among controls in the initial ES PDAC study population (figure 2).

All steps of data preprocessing (filtering and normalisation), model training, predictions and model evaluation were performed using the SIAMCAT R package v1.5.0<sup>79</sup> (<https://siamcat.embl.de/>).

### External validation of the metagenomic classifiers

To assess the disease specificity of the trained models, we obtained predictions for samples from other gut metagenomic datasets (online supplemental table S6) for the full list, including accession numbers). We performed a literature search to identify publicly available datasets of faecal metagenomes in case-control or cohort studies for relevant diseases. For a total set of 25 studies covering 5792 samples across nine disease states, raw sequencing data were downloaded from the European Nucleotide Archive and taxonomically profiled as described above.<sup>35–59</sup>

The trained metagenomic classifiers for PDAC were then applied to each external dataset after applying a data normalisation routine which selects the same set of features and uses the same normalisation parameters (for example, the mean of a feature for standardisation by using frozen normalisation functionality in SIAMCAT) as in the normalisation procedure from the pancreatic cancer dataset. Then, predictions were assessed for disease specificity because high prediction scores for samples from other disease samples would indicate that the classifier relies on general features of dysbiosis in contrast to signals specific to pancreatic cancer, which would not result in elevated false-positive rates on samples from other diseases. For this analysis, the cut-off point for the predictions was set at a false-positive rate of 10% among controls in the initial PDAC study population (figure 3). The effect of age, sex and sequencing depth of 25 populations on prediction score were tested by using the *cor.test* function (Spearman method) in the *car* R package v3.0–3.

### Subspecies and strain-level analyses

Metagenomic reads were mapped against species-representative genomes from the proGenomes v1 database<sup>80</sup> (see above). Microbial single nucleotide variants were called from uniquely mapping reads using metaSNV,<sup>81</sup> and within-species allele distances between samples were calculated as described previously.<sup>82</sup> Associations between allele distance and PDAC disease state were quantified using PERMANOVA after stratifying for potential confounders (including sampled body site).

Oral-intestinal transmission of strains was quantified as described previously.<sup>83</sup> In short, the overlap between microbial single nucleotide variants in salivary and faecal samples within subjects was contrasted with a between-subject background to compute a quantitative oral-faecal transmission score and *p* value. Associations of species- and subject-specific transmission scores with clinical factors were tested using ANOVA and *post hoc* tests, followed by a Benjamini-Hochberg correction for multiple tests.

### Fluorescence *in situ* hybridisation microscopy

FISH analyses were performed using probes specifically targeting the 16S rRNA sequence unique to a particular taxon of bacteria (figure 4). All probes were selected based on a literature search and the corresponding taxa are displayed in online supplemental table S7).

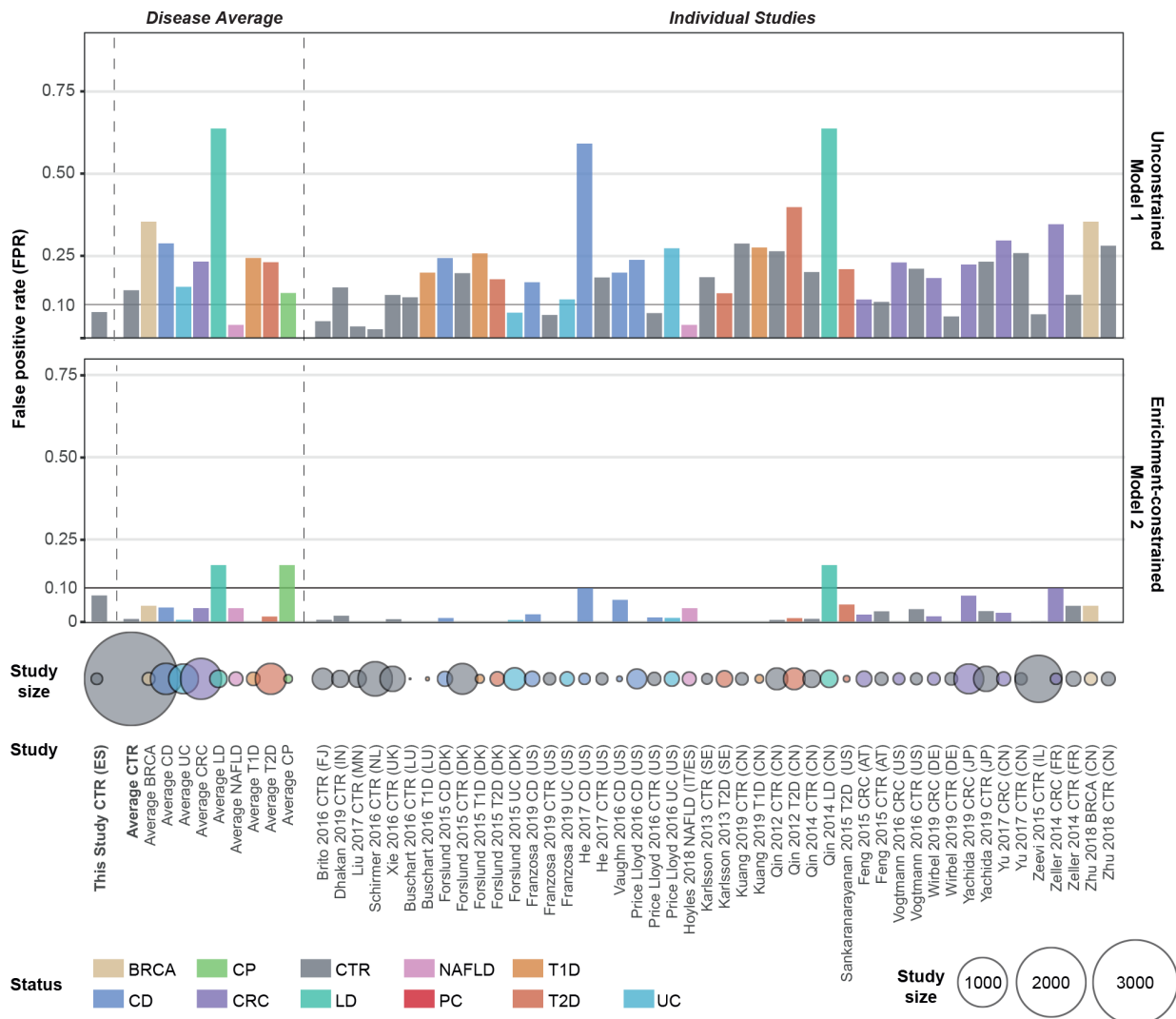
Pancreatic tumour and normal pancreas samples were obtained from the pathology department and immediately frozen in liquid nitrogen within less than 30 min of surgical excision. Sterile material was used to dissect the different samples. The minimum size of tissue for freezing was approximately 0.125 cm<sup>3</sup> (0.5×0.5×0.5 cm). Samples were transferred from the temporary liquid nitrogen transport container and kept in a locked freezer at –80°C. Before analysis they were transported on dry ice, moved to an optimal cutting temperature mould in liquid nitrogen and immediately cut on a cryotome to obtain 10 sections of 3–5 µm each. All material was sterilised with ethanol after each sample handling.

Tissue sections of 5 µm thickness were mounted on positively charged slides (SuperFrost, Thermo Scientific). Briefly, tissues were postfixed in freshly prepared 4% paraformaldehyde. After enhancement of the bacteria wall permeabilisation by lysozyme treatment (10 g/L Tris HCl 6.5M), samples were hybridised for 1 hour at 45°C in the presence of the specific probe in a hybridiser machine (DAKO). Hybridisation was done in 20 µL of hybridisation buffer (20 nM Tris, pH 8.0. 0.9 M NaCl, 0.02% sodium dodecyl sulfate, 30% formamide) added to 100 ng of the probe. Finally, the tissues were washed in washing solution (70% formamide, 10 mM Tris pH7.2 and 01% bovine serum albumin), dehydrated in a series of ethanol samples, air-dried and stained with 0.5 µg/mL DAPI (4',6'-diamidino-2-phenylindole)/antifade solution (Palex Medical). FISH images were captured using a Leica DM5500B microscope with a CCD camera (Photometrics SenSys) connected to a PC running the CytoVision software 7.2 image analysis system (Applied Imaging). Images were analysed blind and scored based on the intensity of the probe signal.

## RESULTS

### PDAC is associated with moderate shifts in microbiome composition when controlling for confounding factors in shotgun metagenomic data

We studied 57 newly diagnosed, treatment-naïve patients with PDAC, 29 patients with chronic pancreatitis (CP), and 50 controls matched for age, gender and hospital. Participants were prospectively recruited from two hospitals in Barcelona and Madrid, Spain, between 2016 and 2018, using the same standards (see subject characteristics in figure 1A and online supplemental table S1–S3 for the clinical data for each subject). We obtained faecal shotgun metagenomes for all subjects and salivary metagenomes for 45 patients with PDAC, 12 with CP, and 43 controls (see 'Methods'). The

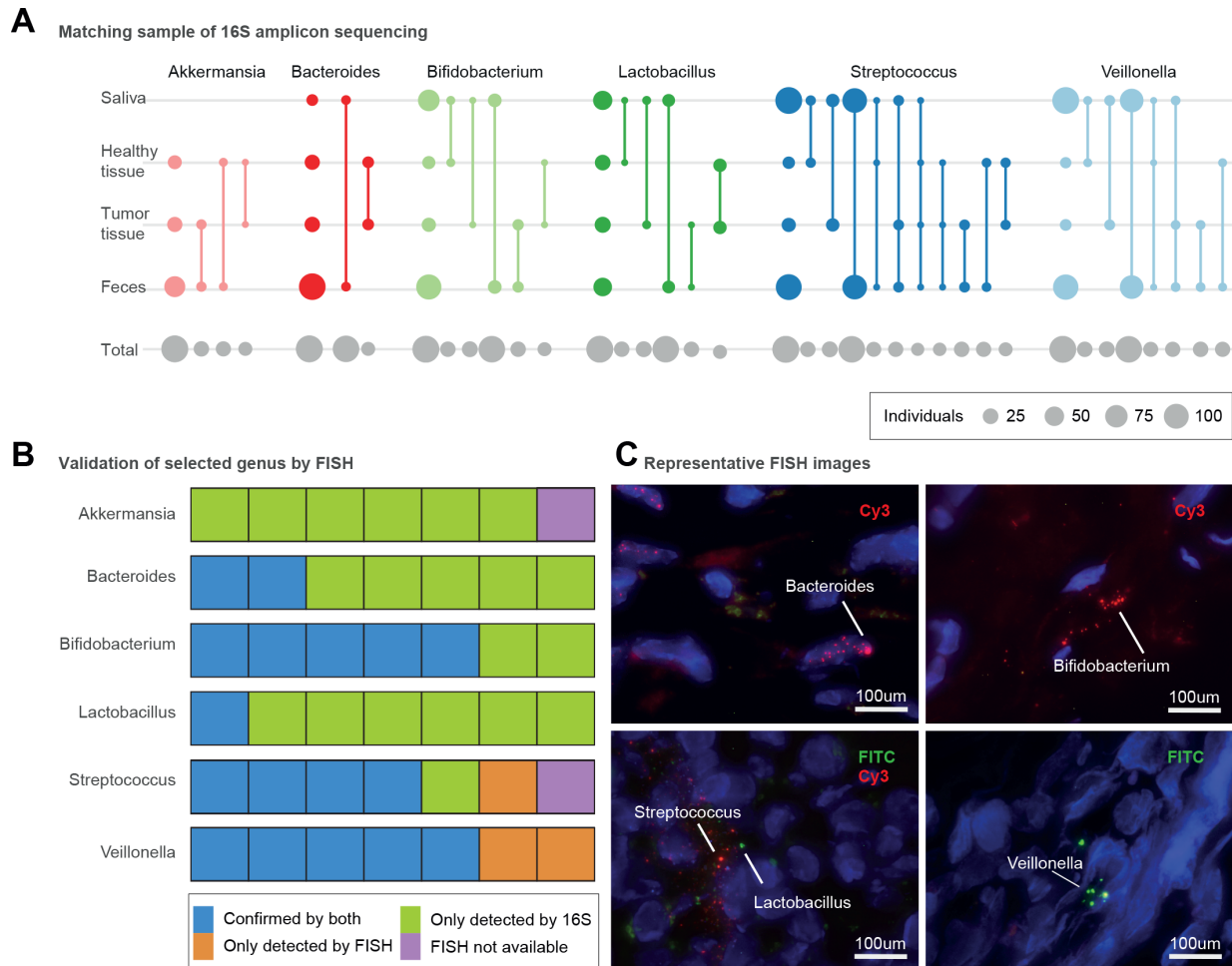


**Figure 3** External validation of the disease specificity of pancreatic ductal adenocarcinoma (PDAC) faecal microbiome models. False positive rate (FPR) of metagenomic unconstrained model-1 and enrichment-constrained model-2 in 25 external test sets is shown as a bar plot (see online supplemental table S4 for a list of all studies included). Validation datasets were profiled and normalised in the same way as the initial dataset (see 'Methods'). Each study was stratified according to health status and models were tested to predict in the given group at a 90% specificity cut-off. A low FPR on metagenomes from patients with other disorders and healthy individuals indicates that the model is specific to PDAC. The number of subjects in each group is displayed as colour coded circles below. BRCA, breast cancer; CRC, colorectal cancer; CD, Crohn's disease; CP, chronic pancreatitis; CTR, controls; LD, liver disease; NAFLD, non-alcoholic fatty liver disease; PC, pancreatic cancer; T1D, type 1 diabetes; T2D, type 2 diabetes; UC, ulcerative colitis; ES, Spanish; DE, German.

analysis workflow is detailed in online supplemental figure 1.

As several PDAC risk factors, such as tobacco smoking, alcohol consumption, obesity or diabetes, are themselves associated with microbiome composition<sup>84</sup>, we first sought to establish potential confounders of microbiome signatures in our study population, in order to adjust analyses accordingly. For a total of 26 demographic and clinical variables, we quantified marginal effects on microbiome community-level diversity (online supplemental table S4). Faecal and salivary microbiome richness (as a proxy for alpha diversity) were not univariately associated with any tested variable, or with PDAC status, when accounting for the most common PDAC risk factors and applying a false discovery rate threshold of 0.05 (online supplemental figure 2, online supplemental table S4).

Microbiome community composition, in contrast, varied with age at diagnosis (PERMANOVA on between-sample Bray-Curtis dissimilarities,  $R^2=0.01$ , Benjamini-Hochberg-corrected  $p=0.03$ ), diabetes ( $R^2=0.01$ ,  $p=0.04$ ) and jaundice status ( $R^2=0.02$ ,  $p=0.009$ ) in faeces, and with aspirin/paracetamol use ( $R^2=0.02$ ,  $p=0.04$ ) in saliva, albeit at very low effect sizes (online supplemental table S5). Even though cases and controls were matched for age and sex, we included these factors as strata for subsequent analyses. Under such adjustment, subject disease status was mildly but statistically significantly associated with community composition in faeces ( $R^2=0.02$ ,  $p=0.001$ ), but not in saliva ( $R^2=0.01$ ,  $p=0.5$ ) (figure 1B, online supplemental figure 3–4, online supplemental table S5). Indeed, the faecal microbiome composition of patients with PDAC differed from that of both controls ( $R^2=0.02$ ,  $p\leq 0.0001$ ) and patients with CP



**Figure 4** Presence of microbiomes in different sections of the pancreas with different conditions. (A) Presence of different genera in four different body sites including faecal, saliva, pancreatic tumour and healthy tissue samples, as inferred by 16S amplicon data. Circle size corresponds to the total number of subjects available for each comparison (grey, bottom row) or with intra-individually matched amplicon sequence variants (coloured); matched sample types are connected by lines. The first column shows the total number of samples per site in which the genus was detected. (B) Seven selected pancreatic tissue samples (five tumour and two non-tumour) to show bacterial presence/absence with both 16S amplicon and fluorescence *in situ* hybridisation (FISH) methods. Validation of bacterial presence with both 16S amplicon sequencing and FISH is shown in blue. Samples showing bacterial presence according to 16S only are displayed in green. Bacterial presence validated only by FISH is shown in orange, and samples not subjected to FISH validation owing to lack of tissue material are shown in purple. (C) Representative microscopy images for *Bacteroides* (intranuclear, tumour tissue), *Bifidobacterium* (extranuclear, tumour tissue), *Lactobacillus* (extranuclear, non-tumour tissue), *Streptococcus* (extranuclear, non-tumour tissue), *Veillonella* (extranuclear, tumour tissue). Fluorescein isothiocyanate (FITC) and Cy3 fluorescent dyes were used as indicated, and DAPI (4',6-diamidino-2-phenylindole; blue) was used to label the nucleus.

( $R^2=0.02$ ,  $p=0.003$ ), although likewise at very small effect sizes.

#### High-accuracy metagenomic classifiers capture specific faecal microbiome signatures in patients with PDAC

Having established the presence of a gut microbiome signal for PDAC at the coarse level of overall community composition, we next identified nine species with disease-specific univariate associations (Wilcoxon test of relative abundances in PDAC cases vs controls, Benjamini-Hochberg-corrected  $p<0.05$ ; see figure 1c). Most prominently, *Veillonella atypica*, *Fusobacterium nucleatum/hwasookii* and *Alloscardovia omnicolens* were enriched in faeces of patients with PDAC, whereas *Romboutsia timonensis*, *Faecalibacterium prausnitzii*, *Bacteroides coprocola* and *Bifidobacterium bifidum* species clusters were depleted. In contrast, we did not detect any species with significantly differential abundance in the salivary microbiome when correcting for

multiple tests, including previously reported associations, such as *Porphyromonas gingivalis*, *Aggregatibacter actinomycetemcomitans*,<sup>22</sup> *Neisseria elongata* or *Streptococcus mitis*<sup>18</sup> (online supplemental figure 5).

Among the univariately associated faecal species, several were by themselves moderately predictive of PDAC state (online supplemental figure 5). To coalesce such individual signals into an overarching model, we next built multispecies metagenomic classifiers by fitting LASSO logistic regression models in 10-fold cross-validation (see 'Methods'). When applying no further constraints, the obtained model discriminated between patients with PDAC and controls with high accuracy in our study population ('model-1'; AUROC=0.84; Figure 2). The most prominent positive marker species in the model were *Methanobrevibacter smithii*, *Alloscardovia omnicolens*, *Veillonella atypica* and *Bacteroides finegoldii*. We note that by design, LASSO regression selects representative features among inter-correlated sets;

therefore, these species may be representatives of larger species sets with highly correlated abundances. None of the 26 demographic and epidemiological variables describing our study population were selected as predictive features by the model, and the microbiome signature was more informative than any other feature (see online supplemental figure 6 and 7). Further, none of these variables were individually associated with the microbial species represented in the model, ruling them out as potential confounders. This indicates that the classifier captured a diagnostic gut microbiome signature of PDAC that is probably independent of other disease risk factors and potential confounders.

An analogous model built to differentiate patients with CP from controls had no predictive power (AUROC=0.5; online supplemental figure 8), consistent with the observation that these groups were compositionally largely indistinguishable. Similarly, no robust PDAC signature was detected for the salivary microbiome (AUROC=0.48; online supplemental figure 9). However, a faecal model to distinguish patients with PDAC from those with CP performed better with an AUC of 0.75, but model robustness was limited by the low sample size in the group with CP (online supplemental figure 8). We further explored predictive associations at the higher resolution of functional microbiome profiles. Models based on the abundances of KEGG modules (online supplemental figure 10) achieved an accuracy of up to AUROC=0.74, but feature selection was likewise not robust across validation folds, as a consequence of fitting a high number of variables (modules) against a limited set of samples. We therefore pursued the species-based classifiers, as they provided stable models.

The initial gut microbiome-based classifier included several species depleted in PDAC relative to controls, such as *Faecalibacterium prausnitzii*, *Bacteroides coprocola*, *Bifidobacterium bifidum* or *Romboutsia timonensis* (figure 2B). For some of these species, it was previously suggested that depletion is linked to intestinal inflammation, in general, rather than to specific diseases.<sup>85</sup> We therefore retrained a classifier with the constraint that positively associated (enriched) microbial features were exclusively selected in each cross-validation fold. The resulting enrichment-constrained model (model-2) discerned patients with PDAC with an accuracy of AUROC=0.71. The difference with the unconstrained model, model-1, was mostly attributable to a penalty on sensitivity—that is, a decrease in confident detections of patients with PDAC, in line with expectations when training on sparse data.

### Combination of metagenomic classifiers with antigen CA19-9 levels increases accuracy

Blood serum levels of the antigen CA19-9 are routinely used to monitor PDAC progress,<sup>86,87</sup> but have also been suggested as a potential marker for early diagnosis of PDAC, although with moderate reported sensitivity (0.80, 95%CI 0.72 to 0.86) and specificity (0.75, 95%CI 0.68 to 0.80).<sup>12</sup> CA19-9 serum levels were available for a subset of 77 individuals (33/50 controls and 44/57 patients with PDAC) in our Spanish population (online supplemental figure S11). Given that CA19-9 is directly secreted by tumours, we hypothesised that the readouts provided by CA19-9 serum levels and by our microbiome classifiers were complementary, and that their combination could improve the accuracy of PDAC prediction. Indeed, accounting for CA19-9 increased the accuracy of our unconstrained model-1 from AUROC=0.84 to 0.94, driven mostly by an increase in sensitivity (figure 2B). More strikingly, when we amended the enrichment-constrained model-2 with CA19-9 information, we observed a large increase in accuracy from AUC=0.71 to 0.89, likewise driven by a significant improvement in sensitivity, thereby essentially abolishing the performance penalty relative to

model-1 (figure 2C, online supplemental figure S11). There was no significant bias towards higher CA19-9 levels in later disease stages in either the ES or DE populations (online supplemental figure S11).

Our Spanish study population included 25 patients with PDAC in early disease stages (T1, T2) and 32 subjects in later stages (T3, T4). Disease stage did not affect the performance of either microbiome-based model (figure 2D); in particular, recall was not biased towards later stages.

### Performance of metagenome-based classifiers generalises to independent validation cohorts

To test whether the observed microbiome signatures generalise beyond our focal Spanish study population, we next challenged our models in two validation scenarios. First, we tested prediction accuracy in an independent study population of 44 patients with PDAC and 32 matched controls, recruited from two hospitals in Erlangen and Frankfurt am Main, Germany (see figure 1, Methods and online supplemental table S3), with the samples being processed identically to those of the Spanish population. On this DE validation population, both the unconstrained model-1 (figure 2B) and the enrichment-constrained model-2 (figure 2C) performed with comparable or indeed superior accuracies to the training population, both with and without complementation by CA19-9 levels, and with similar trends across disease stages (figure 2D).

Next, to confirm that our metagenomic classifiers captured PDAC-specific signatures, rather than unspecific, more general disease-associated variation, we further validated them against independent, external metagenomic datasets on various health conditions. In total, we classified 5792 publicly available gut metagenomes from 25 studies across 18 countries, including subjects with CP (this study), type 1 or type 2 diabetes, colorectal cancer, breast cancer, liver diseases, non-alcoholic fatty liver disease, including Crohn's disease and ulcerative colitis, as well as healthy controls (figure 3 and online supplemental table S6).

When tuned to 90% specificity (allowing for 10% false positive predictions) in our focal ES study population, the unconstrained model-1 showed a recall of 56% of patients with PDAC in the ES population and 48% in the DE validation population (with 6% false-positive rate), and up to 64% when complemented with information on CA19-9 levels (available for 8/32 controls and 43/44 patients with cases in the DE cohort). The disease specificity of model-1, however, was limited, with predictions of PDAC state for 15% of control subjects on average across all external datasets. Most of these false positive calls were observed in two Chinese populations of patients with Crohn's disease<sup>48</sup> or liver cirrhosis.<sup>44</sup> Crohn's disease has been associated with depletion signatures similar to those observed in our model (in particular of *F. prausnitzii*,<sup>88</sup>) whereas liver diseases share some physiological characteristics with impaired pancreas function. However, all other liver disease and Crohn's disease sets showed lower false detection rates, indicating that the effect was probably attributable, in part, to technical and demographic effects between studies. Indeed, we note that subjects in these two Chinese study populations were significantly younger than our populations (50±11 years for Qin\_2014; 28.5±8 years for He\_2017; 70±12 years for our ES population). This age effect was systematic: across all validation sets, PDAC prediction scores were associated with subject age (ANOVA  $p=0.007$ ;  $\rho_{\text{Spearman}}=0.16$ ), as well as with the sex of the subject ( $p<10^{-6}$ ) and sequencing depth ( $p=0.0008$ ;  $\rho_{\text{Spearman}}=0.1$ ) (online supplemental figure S12, online supplemental table S6).

The enrichment-constrained model-2 showed lower detection rates in patients with PDAC in both populations, although recall

was reinstated for CA19-9 combined models. Model-2 was highly specific for PDAC with, on average, just 0–5% PDAC predictions in almost all external populations, at a maximum of 17% predictions among the aforementioned<sup>44</sup> population with liver disease. In particular, the detected microbiome signatures were also robust against misclassification of patients with type 2 diabetes (<2% false-positive rate); this is relevant to potential screening applications, as these patients are a major PDAC risk group (figure 3).

### PDAC harbours characteristic bacteria, consistent with oral and gut microbiome communities

Alterations in pancreatic secretion, as a consequence of tumour growth in the pancreatic duct, can affect digestive function and may thus plausibly underlie characteristic gut microbiome signatures, such as those described above. This would imply that PDAC progression can indirectly cause microbiome shifts (ie, reverse causation). In addition, the pancreatic duct directly communicates with the duodenum, providing an anatomical link for bacteria<sup>25 30 89</sup> and fungi<sup>34</sup> to colonise the pancreas and contribute to carcinogenesis.<sup>31</sup>

We therefore hypothesised that several gut microbial taxa associated with PDAC should be detectable in pancreatic tumours. We taxonomically profiled all faecal and salivary samples, as well as biopsies of tumours (n=23) and adjacent healthy pancreatic tissue (n=20) of patients with PDAC from our study population using 16S rRNA amplicon sequencing, applying strict filters to exclude putative reagent contaminants often seen in samples of low bacterial biomass<sup>33 90</sup> (see ‘Methods’). We observed a surprisingly rich and diverse pancreas microbiome, with at least 13 bacterial genera present in  $\geq 25\%$  of samples, prominently including taxa with characteristic PDAC signatures in the faecal microbiome<sup>91</sup> (figure 4A, online supplemental figure 13). Among these, *Lactobacillus* spp, *Akkermansia muciniphila* and *Bacteroides* spp were enriched in tumours relative to non-tumour pancreatic tissue (Wilcoxon test, false discovery rate-corrected  $p < 0.006$ ).

In a subset of five tumour and two non-tumoural pancreatic tissue samples, we could further verify the prevalence of *Akkermansia* spp, *Lactobacillus* spp, *Bifidobacterium* spp, *Veillonella* spp, *Bacteroides* spp and *Streptococcus* spp using FISH assays with genus-specific primers (online supplemental figure 4, online supplemental table S7). Generally, amplicon and FISH data were concordant, though amplicon-based detection appeared more sensitive probably due to the amount of tissue analysed. Intriguingly, however, *Akkermansia* spp, although observed by amplicon sequencing in 26/30 subjects, were not detectable using FISH in any of the tested samples (figure 4B–C, online supplemental figure 14).

### Links between oral, intestinal and pancreatic microbiomes

We next traced exact amplicon sequence variants (ASVs) across salivary, faecal, tumour and healthy tissue samples within subjects (figure 4A), at the highest taxonomic resolution attainable using 16S rRNA data. *Veillonella* spp, characteristically enriched in stool of patients with PDAC, were highly prevalent in both salivary (100% of subjects) and faecal (87.5%) samples across the entire study population, while oral and faecal types also matched tumour and non-tumour tissue ASVs. Interestingly, we found no intraindividual match in *Veillonella* ASVs between tumour and adjacent tissue samples, indicating that tumor-dwelling *Veillonella* spp may be distinct from those in healthy tissue. In addition, our data confirm previous reports that *Lactobacillus* spp<sup>26</sup> and *Bifidobacterium* spp<sup>25</sup> are present in both PDAC tumour

and non-tumour tissue. For both genera, we found that tumour types corresponded to either oral or faecal ASVs, but not both, whereas no ASVs from healthy tissue were matched with faecal samples, indicating that distinct pancreatic subpopulations may be linked to the mouth and the gut.

Using paired salivary and faecal shotgun metagenomes, we further confirmed that strains of faecal PDAC-associated microbes may be sourced from the oral cavity (online supplemental results).

### DISCUSSION

Early detection of PDAC remains a formidable challenge, at the heart of ongoing efforts to mitigate the burden of this cancer. Currently, the sole FDA-approved biomarker for PDAC is serum CA19-9, mostly used for disease monitoring rather than screening, due to inherent limits of sensitivity and specificity: CA19-9 levels can be elevated in several conditions unrelated to pancreatic cancer, while subjects lacking the Lewis-A antigen do not produce CA19-9 at all.<sup>10–12</sup> Small-scale studies have proposed PDAC markers based on pancreatic tissue,<sup>5</sup> urine<sup>6 7</sup> and blood serum<sup>8 9</sup> with limited applicability. Yet there are currently no screening tools for PDAC in the clinic—in particular, for early disease stages.

In a prospectively recruited study population of newly diagnosed, treatment-naïve patients and matched controls for whom oral, faecal and tissue microbiomes were analysed (figure 1A), we developed metagenomic classifiers that robustly and accurately predict PDAC solely based on characteristic faecal microbial species (figure 2). PDAC signatures captured by our multispecies models were orthogonal to well-established PDAC risk factors (figures 1B and 2A). This suggests that, in practice, the faecal microbiome may be used to screen for PDAC, complementary to other testable markers, with added diagnostic accuracy in combined tests, as has been proposed for colorectal cancer.<sup>39</sup> Indeed, a combination of our microbiome classifiers with CA19-9 data, available for a subset of our population, significantly enhanced the accuracy of PDAC detection (figure 2B–D).

Previous studies have explored links between PDAC and the oral<sup>18–22 26 92 93</sup> or faecal<sup>23 24</sup> microbiome at the limited taxonomic resolution of 16S rRNA sequencing, but provided conflicting reports regarding the association patterns of individual taxa, probably due to heterogeneous experimental and analytical approaches. The non-availability of raw sequence and patient-level clinical data for several PDAC datasets has made comparisons between studies challenging, and thus a consensus on PDAC-associated microbiome signatures has so far failed to emerge. Several previously reported univariate PDAC associations of oral taxa including *P. gingivalis*, *A. actinomycetemcomitans*, *S. thermophilus* and *Fusobacterium* spp were not confirmed in our study population (online supplemental figure 4); we generally did not observe any salivary PDAC signature either for individual species or for multispecies models.

We carefully checked our analyses for demographic, lifestyle, and clinical confounders, as these can show stronger microbiome associations than disease states.<sup>84</sup> We moreover validated our metagenomic classifiers against the independently sampled, yet consistently processed, DE population (figure 2B–D) and against external populations of various health states from 25 different studies (n=5792)<sup>35–59</sup> (figure 3). Both confounder control and external validation are essential when assessing the disease specificity of predictive models, in particular for diseases

like PDAC with low incidence in the general population. This was confirmed in our analyses: among our two metagenomic classifiers, model-1 showed a high accuracy of AUROC=0.84 in our ES study population, driven by a high recall of patients with PDAC. However, model-1 showed only limited disease specificity in external validations, capturing non-specific species depletion signals discriminative between cases and controls in our population, but also shared by subjects with other diseases. These included generic inflammation signatures—for example, a depletion of *F. prausnitzii*, *E. rectale* or *B. bifidum*. Published metagenomic classifiers for various diseases, and in particular previously reported signatures for PDAC, share similar limitations: highly tuned accuracy on the focal population, but non-specific features shared with other diseases. This lack of specificity limits their translation into clinical practice. In contrast, our model-2, constrained to PDAC-enriched features, achieved only moderate accuracy within our populations (AUC=0.71 on ES, AUC=0.85 on DE) due to a penalty on sensitivity, but was highly PDAC-specific with very low false prediction rates in external populations, including known PDAC risk groups such as those with type 2 diabetes. In particular, PDAC-enriched features in both model-1 and model-2 showed little overlap with characteristic faecal microbiome features for other cancer types, such as colorectal cancer, indicating that a combination of our microbiome models with CA19-9 levels (highly sensitive, but not specific to PDAC) is promising. We note that the residual false positive rate among external populations may partly be due to technical heterogeneity, as all external populations were sampled and processed using independent protocols, and that univariate PDAC associations of individual species may be informative, but not disease-specific (Supplementary Discussion). The panel of PDAC-enriched species in model-2 thus shows potential for microbiome-based PDAC screening, given that a combination with complementary information on serum CA19-9 significantly increased accuracy (AUC=0.89 and 0.92).

Our models showed comparable performance across PDAC disease stages, with no bias towards later stages (figure 2B–D). This indicates that characteristic microbiome signatures emerge early during progression of the disease and that the faecal microbiome can serve for the early detection of PDAC.

Our data are strictly observational and cross-sectional. Nevertheless, there are strong indications that the identified faecal microbiome shifts are not merely a consequence of impaired pancreatic function or systemic effects thereof, although indirect effects cannot be ruled out. Several taxa could be traced between the gut and pancreas, with univariate enrichment in tumours relative to adjacent healthy tissue, indicating direct associations of PDAC with the gut microbiome. We confirmed previous observations<sup>25 30 31 89 91</sup> that the human pancreas harbours a microbiome, both by amplicon sequencing, and by FISH for the most comprehensive panel of taxa to date (figure 4). Pancreatic tissue and tumours contain only low bacterial biomass and are therefore prone to contamination in 16S rRNA amplicon data<sup>33</sup>, whereas FISH testing requires specific hypotheses, so a comprehensive cataloguing of the healthy and diseased pancreatic microbiome composition is still emerging. In our study, we carefully filtered our dataset against known kit contaminants and confirmed the presence of various key genera using FISH assays. We moreover observed an intraindividual overlap of exact amplicon sequence variants between oral, faecal and tissue samples, confirming a shared presence across multiple sites for several species at the highest attainable taxonomic resolution for amplicon data.

Faecal populations of characteristic PDAC-associated taxa could thus be traced back to pancreatic tumours. Similarly, we observed significantly increased levels of oral-intestinal strain transmission in patients with PDAC, in particular of PDAC signature taxa, indicating that these may be sourced intraindividually, from the oral cavity (online supplemental results). These findings suggest that the oral, intestinal and pancreatic microbiomes may be intricately linked, and that multibody site study designs such as presented here will be necessary to disentangle their respective roles and interactions in PDAC aetiology.

In summary, the described faecal microbiome signatures enabled robust metagenomic classifiers for PDAC detection at high disease specificity, complementary to existing markers, and with potential towards cost-effective PDAC screening and monitoring. Furthermore, in view of previous reports on microbe-mediated pancreatic carcinogenesis in murine models and humans,<sup>25 30 94</sup> we believe that the presented panel of PDAC-associated bacterial species may be relevant beyond their use for diagnosis, providing promising future entry points for disease prevention and therapeutic intervention.

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**Patient consent for publication** Not applicable.

**Ethics approval** Participants were prospectively recruited from the Hospital Ramón y Cajal in Madrid and Hospital Vall d'Hebron in Barcelona, Spain. Institutional review board ethical approval (CEI PI 26 2015-v7) and written informed consent was obtained from participating centres and study participants, respectively. An independent validation population was recruited at the Department of Surgery, University Hospital of Erlangen (32 PDAC and 32 control samples) and Section for

Translational Hepatology, Department of Internal Medicine I, Goethe University Clinic Frankfurt (12 PDAC samples). The study was approved by the local ethics committees (SGI-3-2019, 451\_18 B). Clinical data, including disease stage and follow-up data, were retrieved from the clinical records of the hospital charts of the respective patients.

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**Data availability statement** Data are available in a public, open access repository. All data relevant to the study are included in the article or uploaded as supplementary information. The raw sequencing data for the samples are made available in the European Nucleotide Archive (ENA) under the study identifiers PRJEB38625 and PRJEB42013. Metadata for these samples are available as Supplementary Tables S1 and S2. Filtered taxonomic and functional profiles used as input for the statistical modelling pipeline are available in Supplementary Data S1 and S2. Analysis code and results available under <https://github.com/psecekartal/PDAC.git>.

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## 1 SUPPLEMENTARY RESULTS & DISCUSSION

### 2 Several PDAC-associated species in the gut may be sourced from the oral cavity.

3 Many microbial species traverse the gastrointestinal tract to form overlapping populations  
4 between the oral cavity and intestine, with increased levels of intra-individual strain transmission  
5 associated with diseases such as CRC (83). Indeed, several prominent marker taxa showing fecal  
6 enrichment in PDAC are common oral commensals, such as *Veillonella sp.*, *Streptococcus sp.* or  
7 *Fusobacterium sp.*. We hypothesized that intestinal populations of these PDAC-associated  
8 species were primarily of oral origin, with generally enhanced levels of autologous oral-intestinal  
9 strain exchange in PDAC patients. Therefore, we explored microbiome links between body sites  
10 at the highest taxonomic resolution attainable with metagenomic data, at the level of strain  
11 populations.

12 We quantified oral-to-gut transmission based on the intra-individual overlap of microbial Single  
13 Nucleotide Variants (SNVs) for species prevalent in both mouth and gut metagenomes, as a proxy  
14 for oral and intestinal strain populations (see Methods, **Supplementary Fig. 15**). We found that  
15 viewed across all subjects and species, PDAC was associated with increased levels of oral-  
16 intestinal strain population overlap (Cohen's  $d = 0.33$ ; ANOVA  $p < 10^{-3}$  when adjusting for species-  
17 level effects and technical, demographic and clinical variables). This observation extended to  
18 individual PDAC-associated species, with enhanced levels of autologous transmission in several  
19 *Veillonellaceae sp.* (*V. dispar*,  $d=0.71$ ; *V. atypica*,  $d=0.6$ ; *V. parvula*,  $d=0.2$ ; *Megasphaera*  
20 *micronuciformis*,  $d=2.47$ ) and *Streptococcus sp.* (*S. salivarius*,  $d=0.51$ ; *S. vestibularis*,  $d=0.49$ ; *S.*  
21 *parasanguinis*,  $d=0.36$ ). The situation was more nuanced among *Bifidobacteriaceae sp.*, with  
22 enhanced transmission in *B. longum* ( $d=2.16$ ) and *A. omnicolens* ( $d=1.24$ ), but less strain overlap  
23 in *B. dentium* ( $d=-0.89$ ). However, due to limits in metagenomic coverage and species prevalence,  
24 our dataset size did not provide sufficient statistical power to significantly discern these trends for

25 individual species with confidence, in particular when adjusting for putative confounders and  
26 correcting for multiple tests. Nevertheless, our data indicates that PDAC patients showed overall  
27 enhanced levels of oral-intestinal transmission, and that intestinal strain populations of PDAC  
28 signature species may be sourced autologously from the oral cavity.

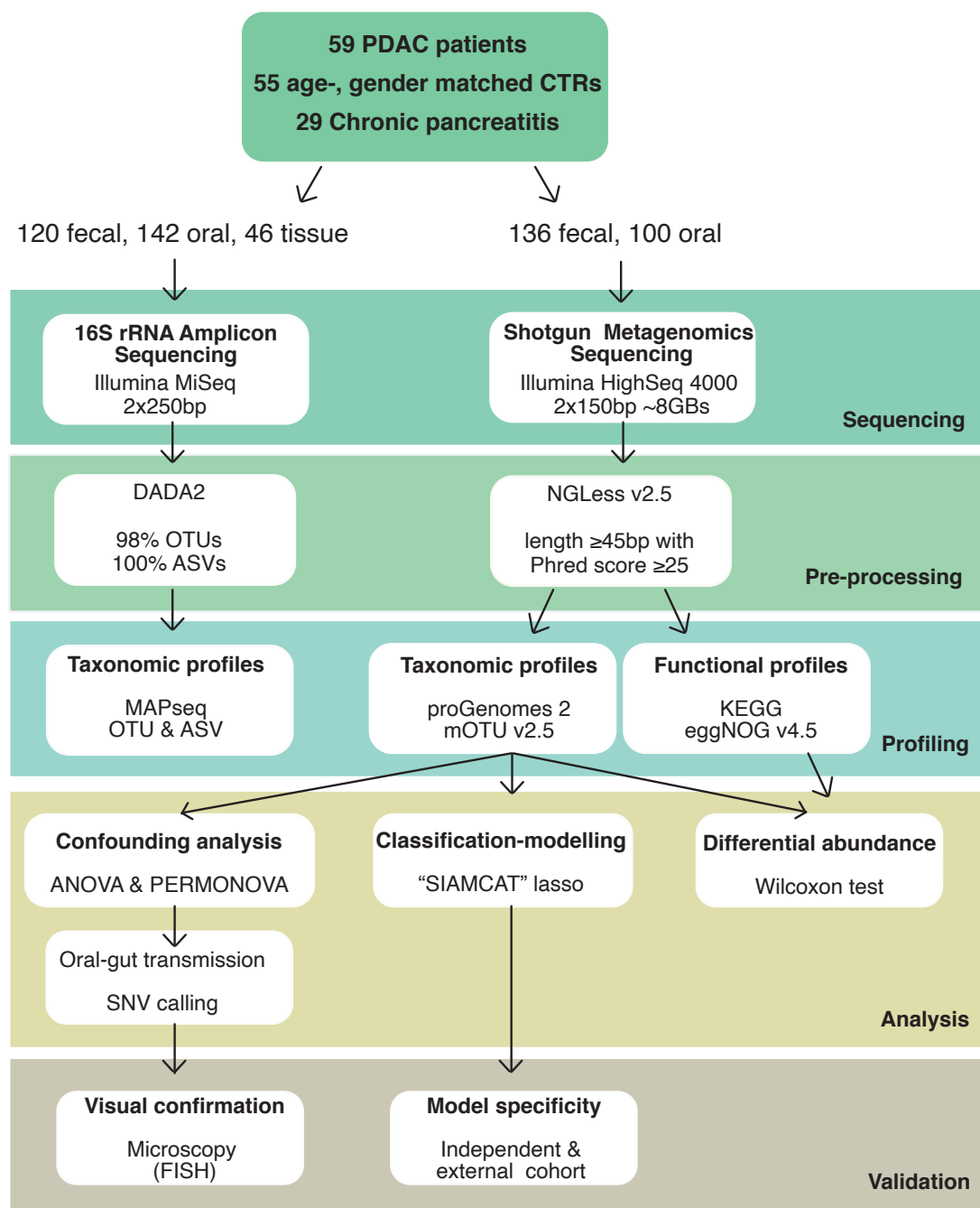
29 **False positive PDAC detections in external validation populations may be due to technical**  
30 **artefacts.**

31 We note that for both model-1 and model-2, at least some false predictions in external validation  
32 sets may be attributable to technical artefacts: technical variation between studies often exceeds  
33 biological differences in microbiome composition (96), while shallower metagenomic sequencing  
34 depths skew taxonomic profiles and bias against lowly abundant species. Moreover, by design,  
35 the external validation sets were matched for neither age nor sex, and information on clinical  
36 variables with relevance to PDAC was usually not collected or not publicly available. The highest  
37 false detection rates were observed among populations with much younger subjects than would  
38 normally be considered a PDAC risk group (**Supplementary Fig. 12**). To overcome such  
39 limitations, meta-studies of multiple geographically and ethnically diverse PDAC cohorts will be  
40 required to further establish globally consistent PDAC microbiome signatures, as has been  
41 successfully shown for colorectal cancer (38,97).

42 **Univariate associations of individual species may be informative, but not specific to PDAC.**

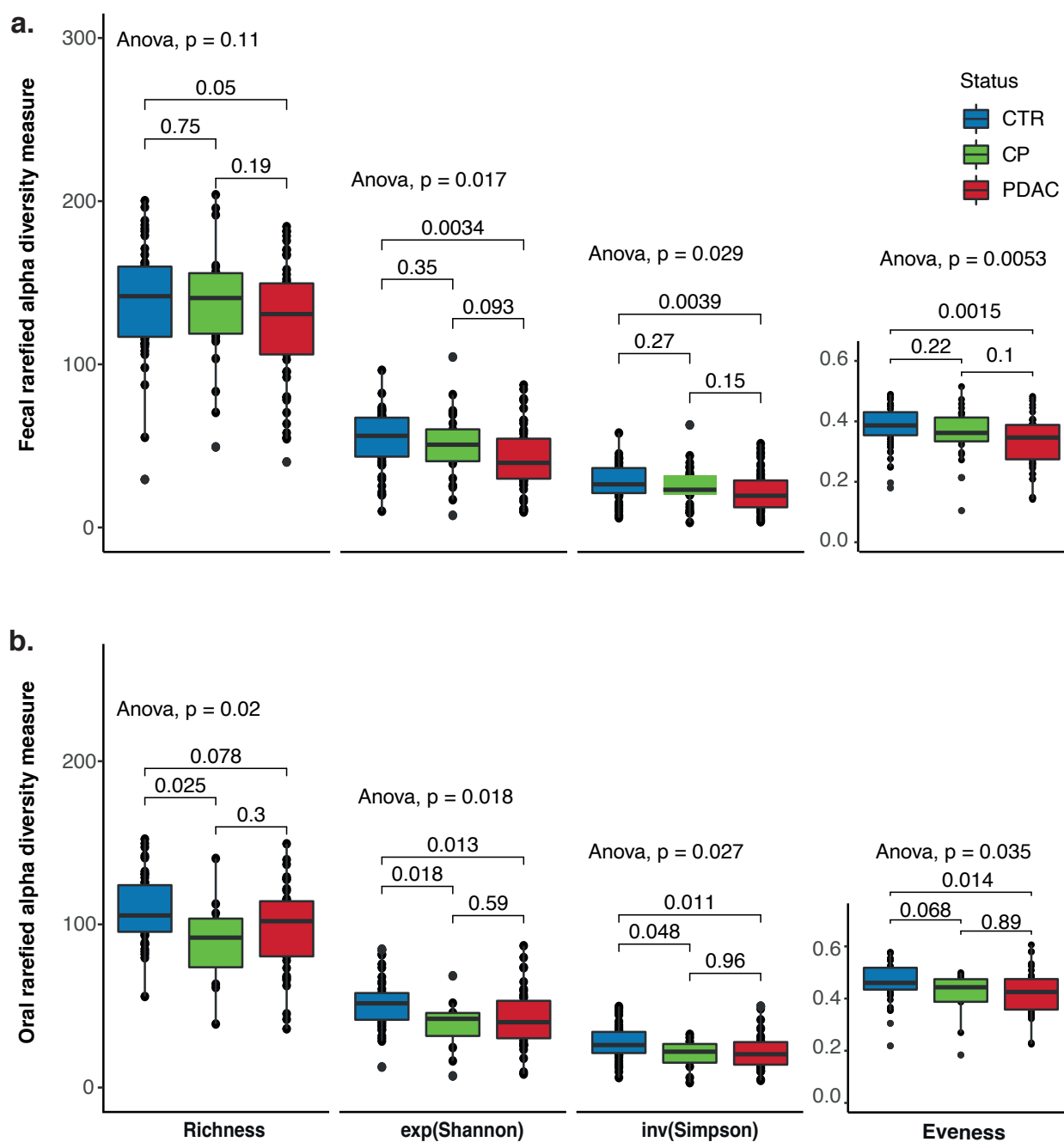
43 Species enriched in PDAC included various *Veillonella sp.*, *Alloscardovia omnicoles*, and  
44 *Methanobrevibacter smithii*, among others (**Fig. 1c and Fig. 2a**). We confirmed that these were  
45 generally not univariately associated with putative confounding factors (**Supplementary Fig. 7**),  
46 yet we note that several among them have previously been linked to both health and disease. For  
47 example, *Veillonella sp.* are common oral and gut commensals and have been associated with  
48 exercise performance in athletes (98), but also with various disease states including cystic fibrosis

49 (a PDAC risk factor) (99), several infections including meningitis (100), as well as lung (101) and  
50 oral carcinomas (102). The role of *Methanobrevibacter smithii*, a prevalent methanogenic  
51 archaeon, in the human gut remains poorly understood (103,104), but the species has likewise  
52 been associated with athletic performance (105) and disease states (104) such as anorexia  
53 nervosa (106,107) and irritable bowel disease (108). This indicates that individual univariate  
54 species associations may be informative, but not specific to PDAC. In contrast, our multi-species  
55 classifier model-2, capturing a combined signature of PDAC-enriched species, provided very high  
56 disease specificity.



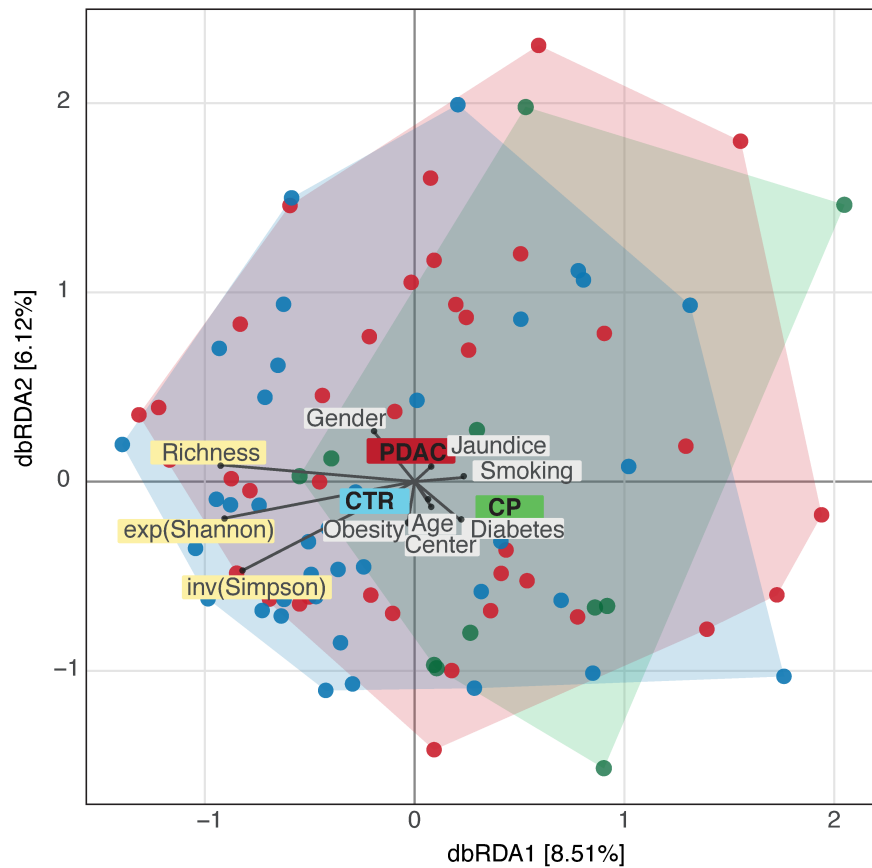
**Figure S1. Analysis workflow.**

Diagram of analysis steps for 16S rRNA amplicon sequencing data and for shotgun metagenomics sequencing data.



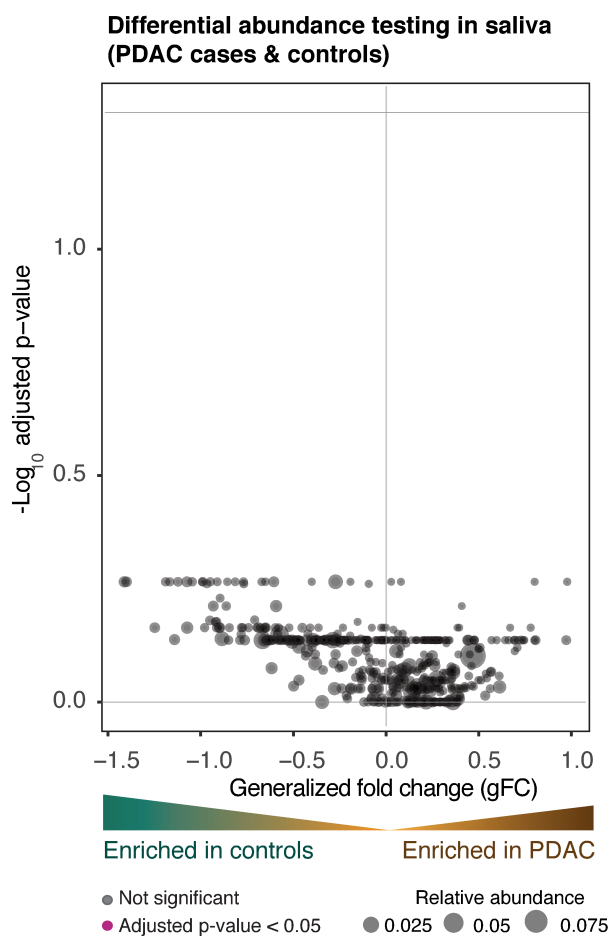
**Figure S2. Alpha diversity measurements comparing PDAC and CP patients with controls.**

Alpha diversity metrics for (a) fecal and (b) oral samples calculated as richness, exponential Shannon index (exp(Shannon)), inverse Simpson index (inv(Simpson)) and eveness. Colors denote groups, with blue for controls (CTR), green for chronic pancreatitis (CP) patients and red for PDAC cases. Pairwise comparisons were performed using Wilcoxon test and comparisons across all three groups were performed using ANOVA (see Methods).



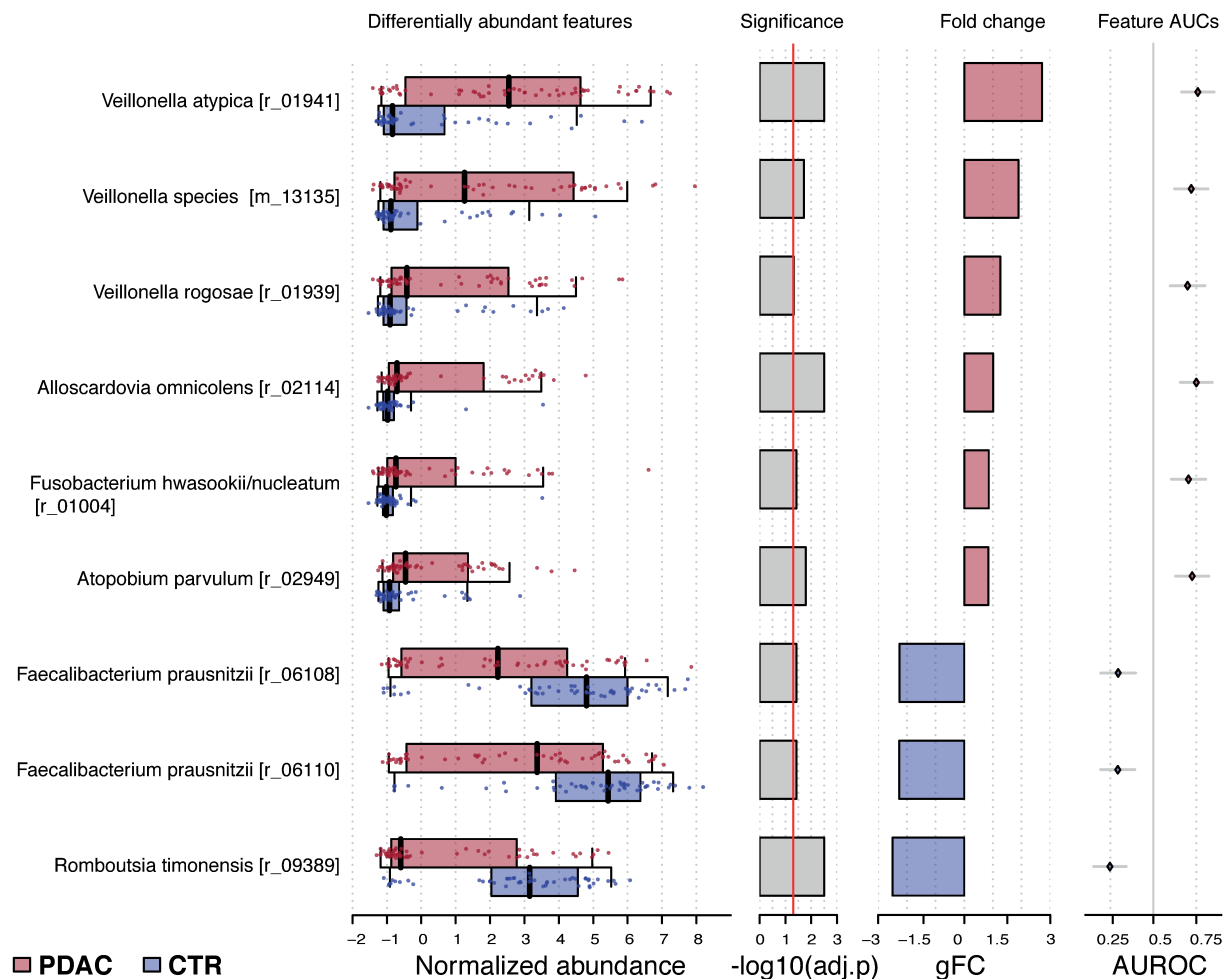
**Figure S3. Distance-based redundancy analysis of saliva microbiome.**

Bray-Curtis distance-based redundancy analysis (dbRDA) of PDAC, CP and control saliva microbiome data. PDAC samples are shown as red circles, CP patients as green and controls as blue. Association with metadata variables are shown as labeled lines. Richness, exponential Shannon ( $\exp(\text{Shannon})$ ) and inverse Simpson ( $\text{inv}(\text{Simpson})$ ) diversity measures are also visualized with lines and were analysed similarly to metadata variables. The length of the metadata variable line represents the confounding effect size (see Methods).



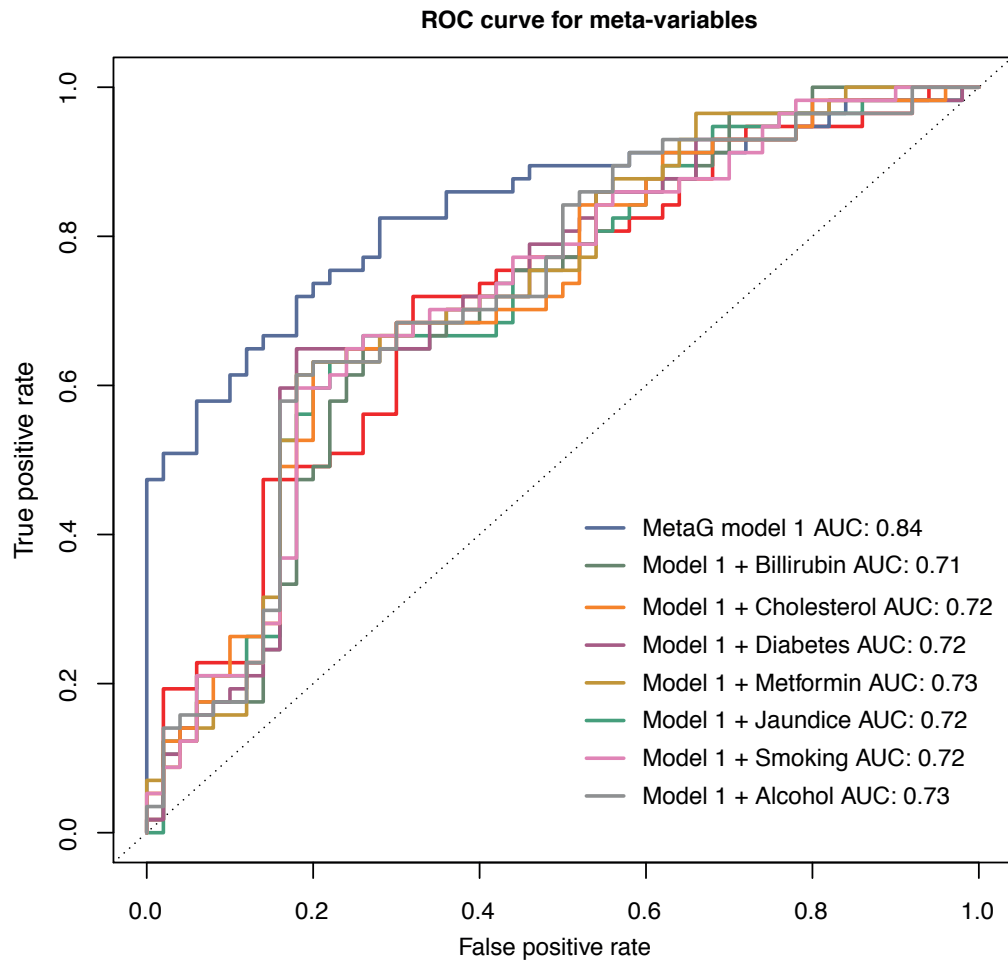
**Figure S4. Differential abundance testing of saliva microbiome**

Wilcoxon test results of saliva microbiome data to test for enrichment of taxa between PDAC cases and controls (see Methods). Y-axis is  $\log_{10}$ (FDR corrected p-values), x-axis is generalized fold change and dot size represents the relative abundance of given species and strains. Red dots represent significantly differentially abundant species/strains in either group, while black dots show non-significant species after FDR correction.



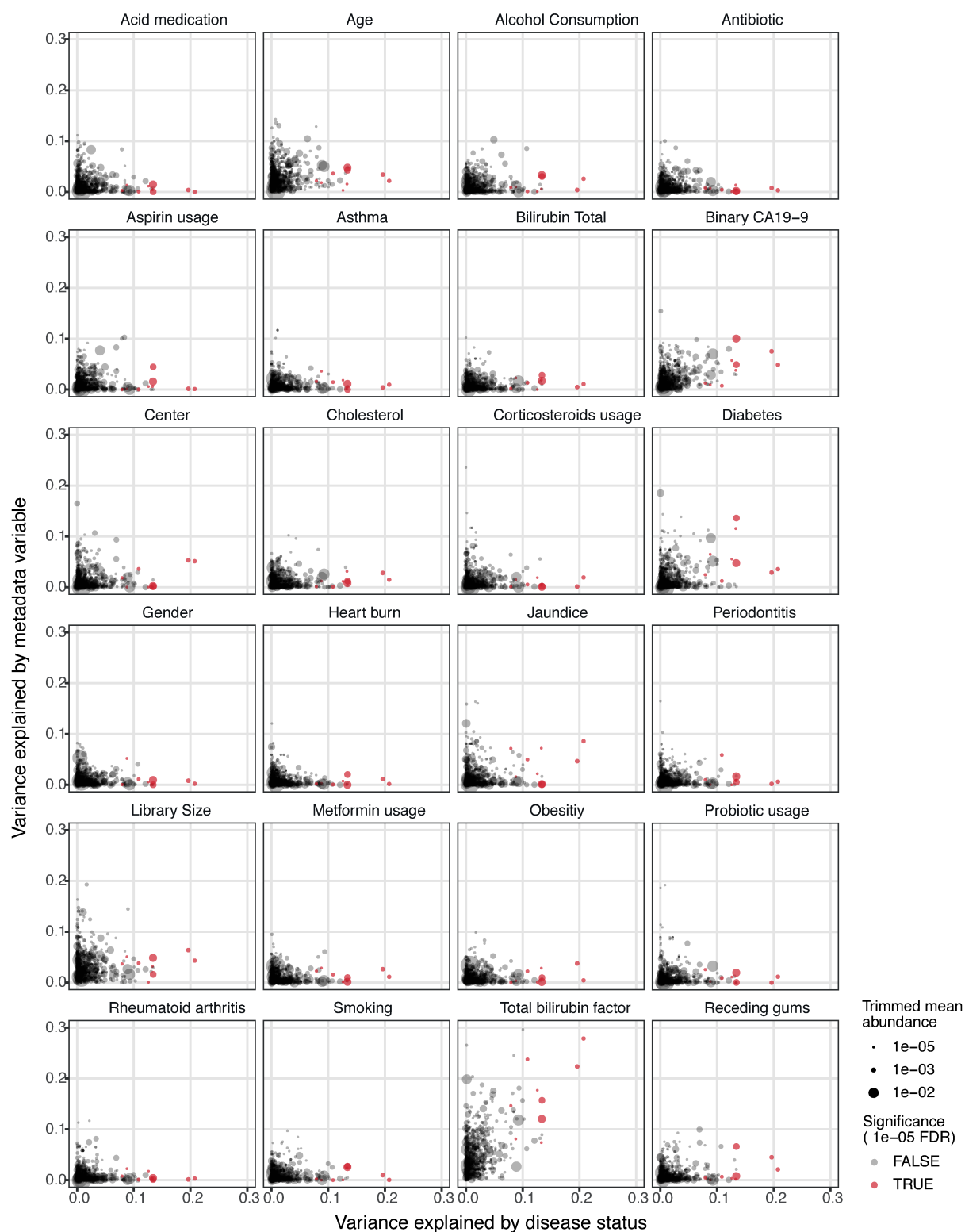
**Figure S5. Differentially abundant species in fecal microbiome between PDAC cases and controls.**

First column panel shows the differentially abundant species between PDAC cases (red) and controls (blue). Middle panels display the  $\log_{10}$ (FDR corrected p-values) and generalized fold change for each taxon and the last panel presents the AUC of each feature to distinguish cases from controls. gFC:Generalized fold change.



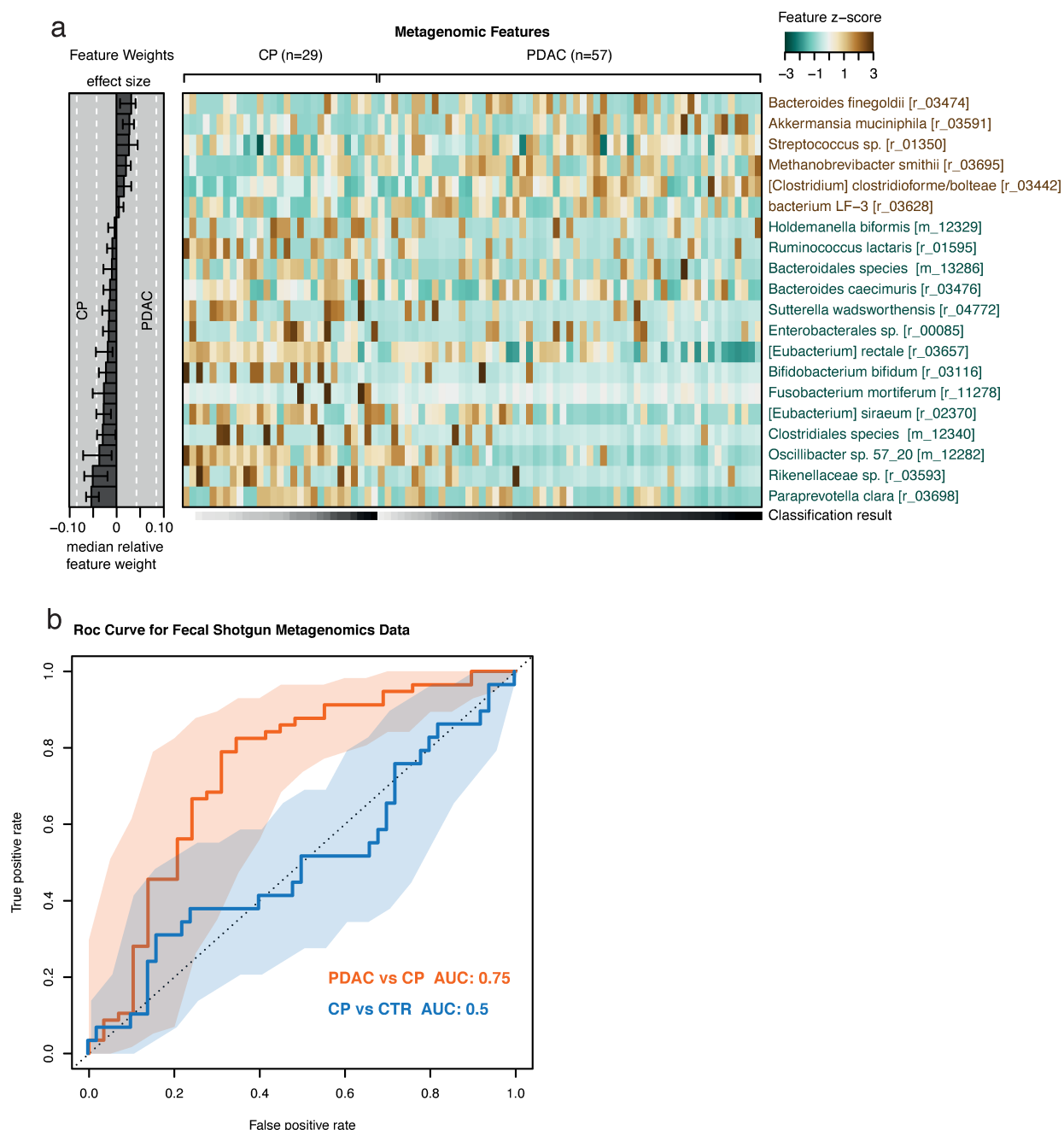
**Figure S6. Contribution of confounding factors to the model.**

The area under the ROC curve (AUROC) is used to show the performance of lasso\_II model based on fecal microbiome data of PDAC and control samples with 10 times resampling and 10 cross validation (see Methods). Each color corresponds to one specific model based on metagenomics features with an additional metadata variable. Shown metadata variables were added to the metagenomics features table with “add.meta.pred” function from “SIAMCAT” package v1.5.0.



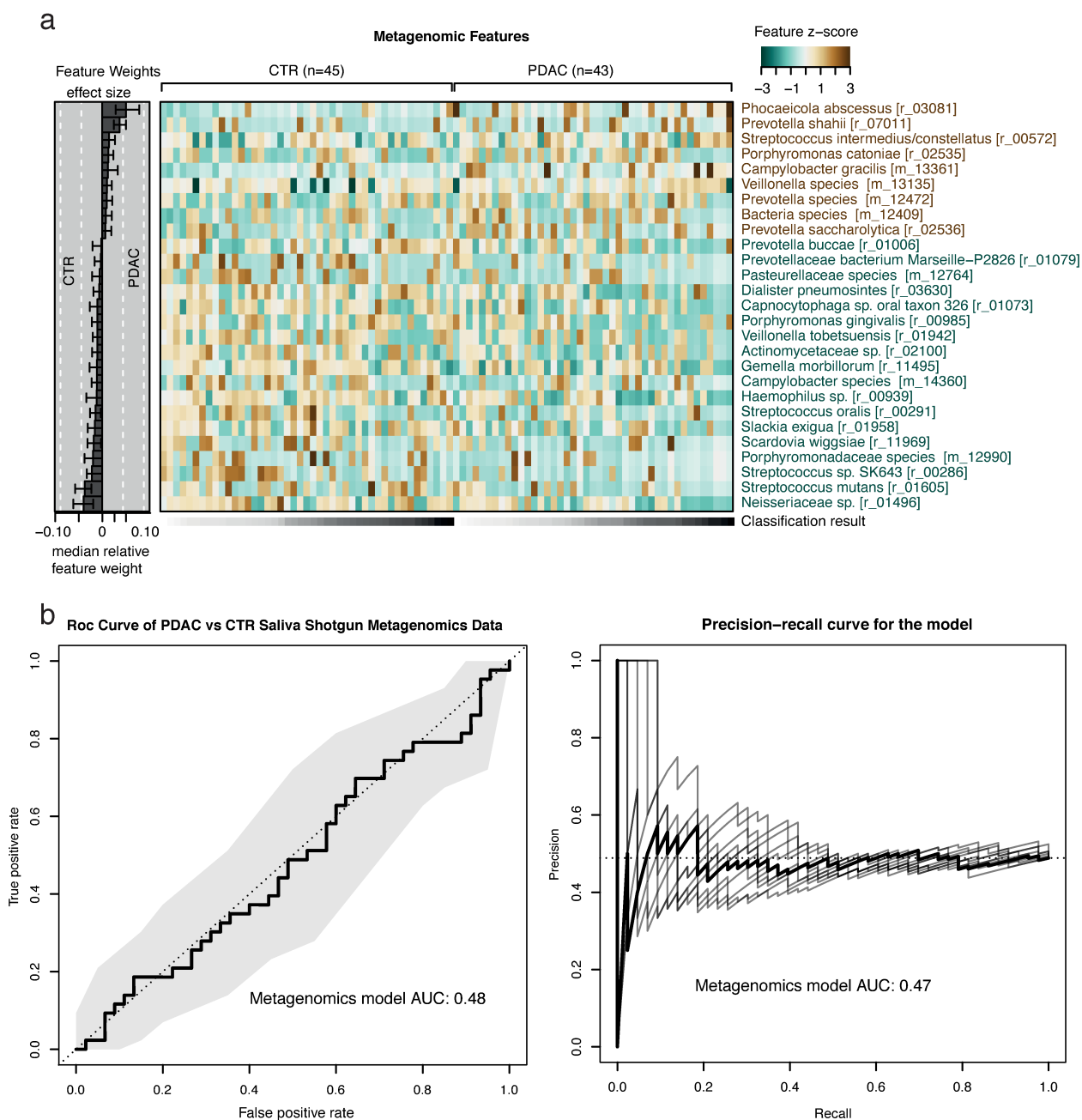
**Figure S7. Potential confounder of single species associations by individual demographic and technical variables.**

Variance explained by diagnosis is represented against confounding factors for single microbial species. Each circle is a strain or species and is colored red if it is differentially abundant between PDAC cases and controls. The size of each circle represents the mean abundance of that species or strain. Disease status and the tested variables were used as explanatory variables in the linear model for feature abundance.



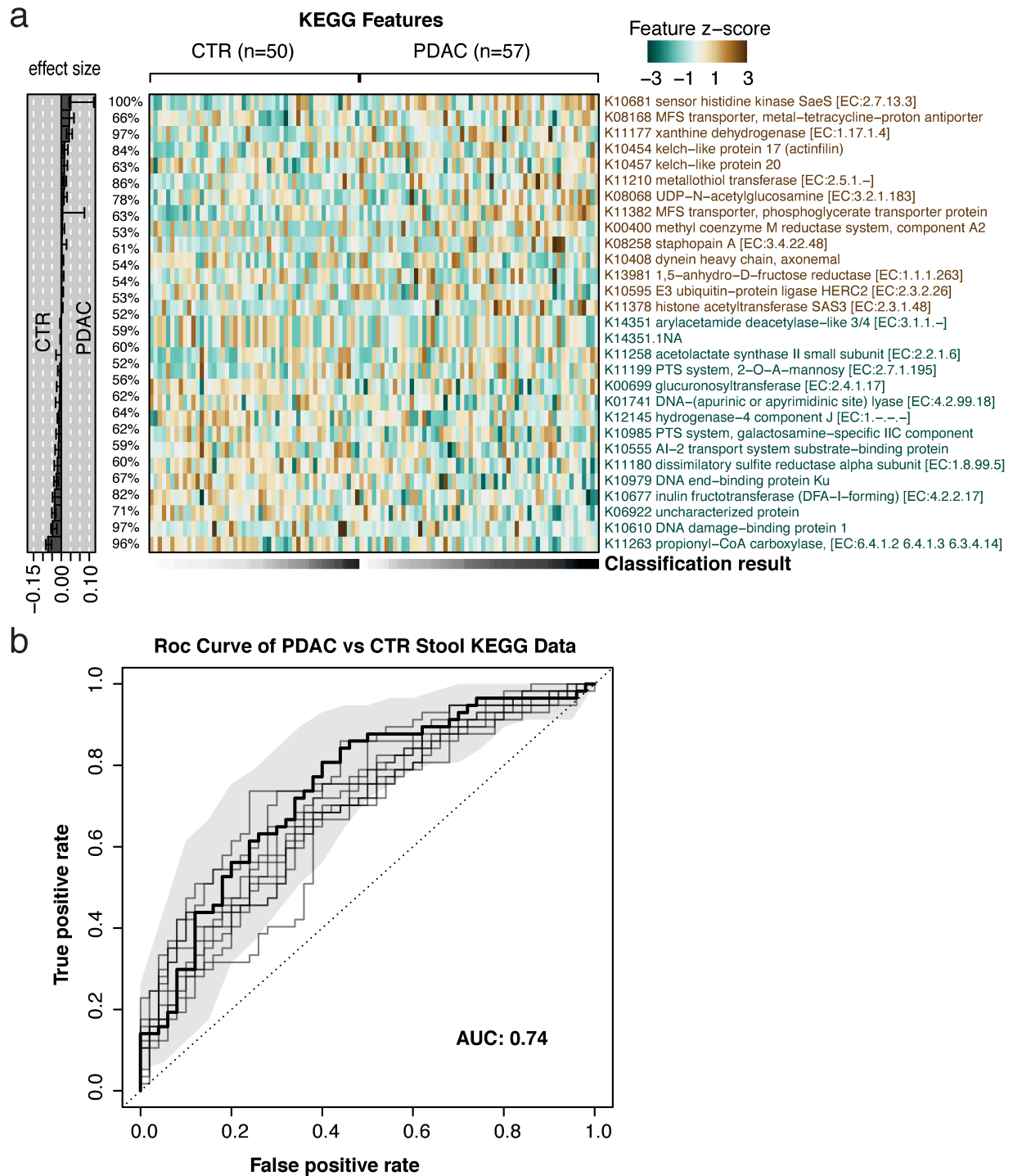
**Figure S8. Fecal microbiome-based classifier distinguishes chronic pancreatitis cases from PDAC patients**

(a) Heatmap representing the selected metagenomic features in the lasso<sub>ll</sub> regression model between PDAC cases and chronic pancreatitis (CP) patients in the fecal microbiome data. (b) ROC curve based on 10 resamplings and 10-fold cross validation (see methods). The blue line represents the model for CP versus controls and the orange line for PDAC vs CP cases. Internal cross validation results are shown as receiver operating characteristic (ROC) curve with a 95% confidence interval shaded in corresponding color.



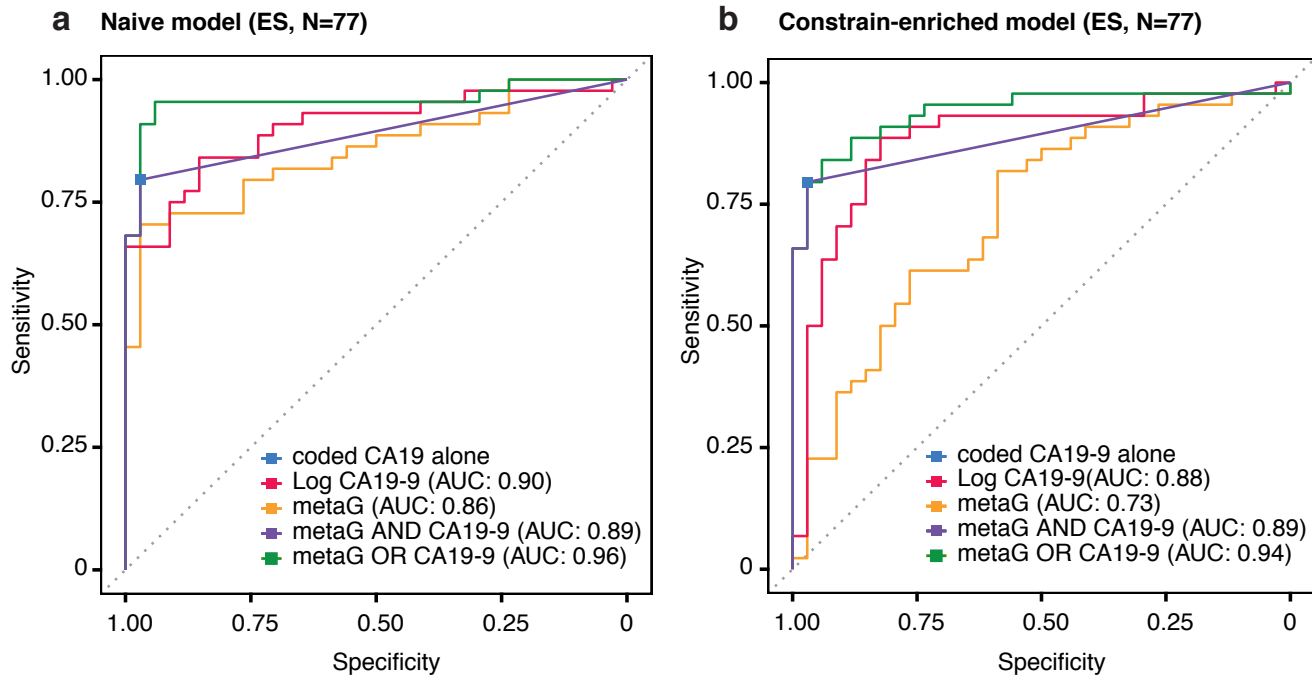
**Figure S9. Oral microbiome does not distinguish PDAC samples from control samples.**

(a) Heatmap representing the selected metagenomic features in the lasso\_II regression model between cases and controls in the saliva microbiome data. (b) ROC curve based on 10 resamplings and 10-fold cross validation (see methods) and precision recall curve. Internal cross validation results are shown as receiver operating characteristic (ROC) curve with a 95% confidence interval shaded in grey.



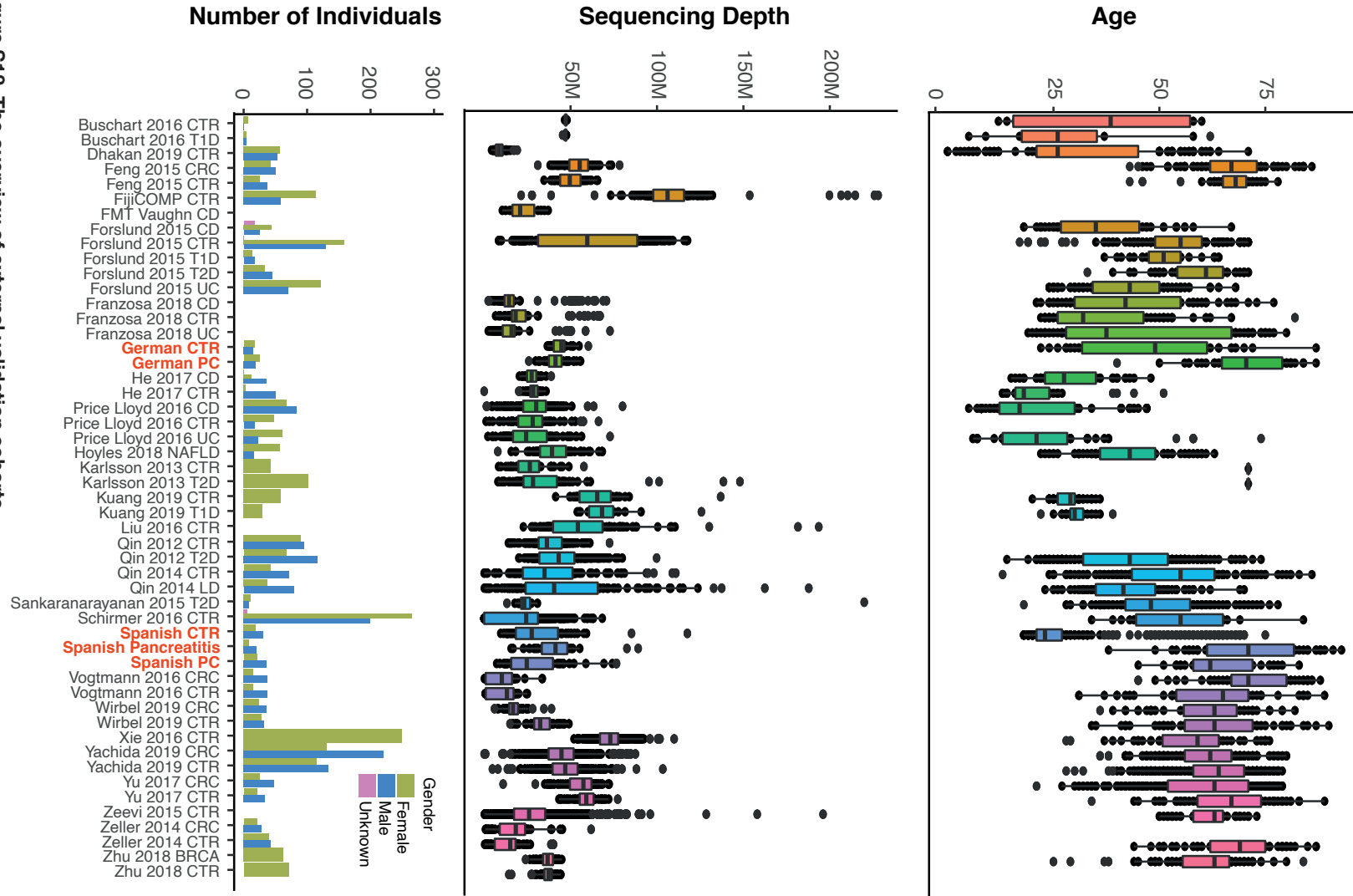
**Figure S10. Lasso\_II regression model based on top 200 KEGG modules.**

(a) Heatmap representing the selected KEGG modules in the lasso\_II regression model. (b) ROC curve based on 10 resamplings and 10-fold cross validation (see methods).

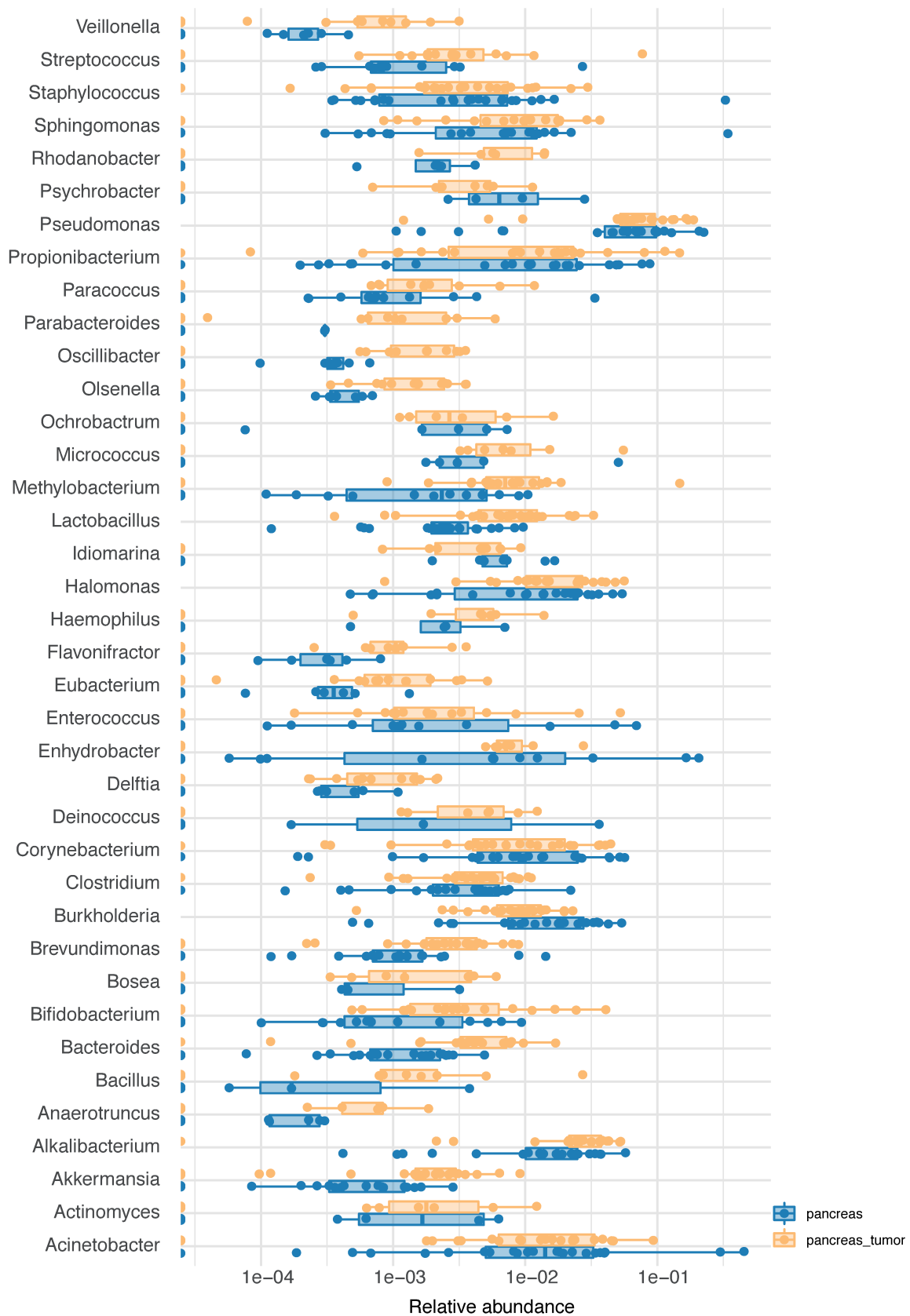


**Figure S11. Combination of fecal microbiome data with CA19-9 results increase sensitivity.**

77/107 (33/50 CTRs and 44/57 PDAC cases) individuals in Spanish (ES) whom CA19-9 data were available included in the modelling process explicitly. CA19-9 values were converted to binary values ( $>37\text{ul/ml} = 1$  &  $<37\text{ul/ml} = 0$ ) (a) ROC curve of full feature set. (b) ROC curve of enrichment-constrained models based on 77 individual fecal microbiomes. Coded CA19-9 is the binary version of data, which is represented by a blue dot. Log(CA19-9) is displayed with red, while “AND” and “OR” combinations are shown with purple and green respectively. 8/32 CTRs and 43/44 PDAC patients in the German (DE) cohort

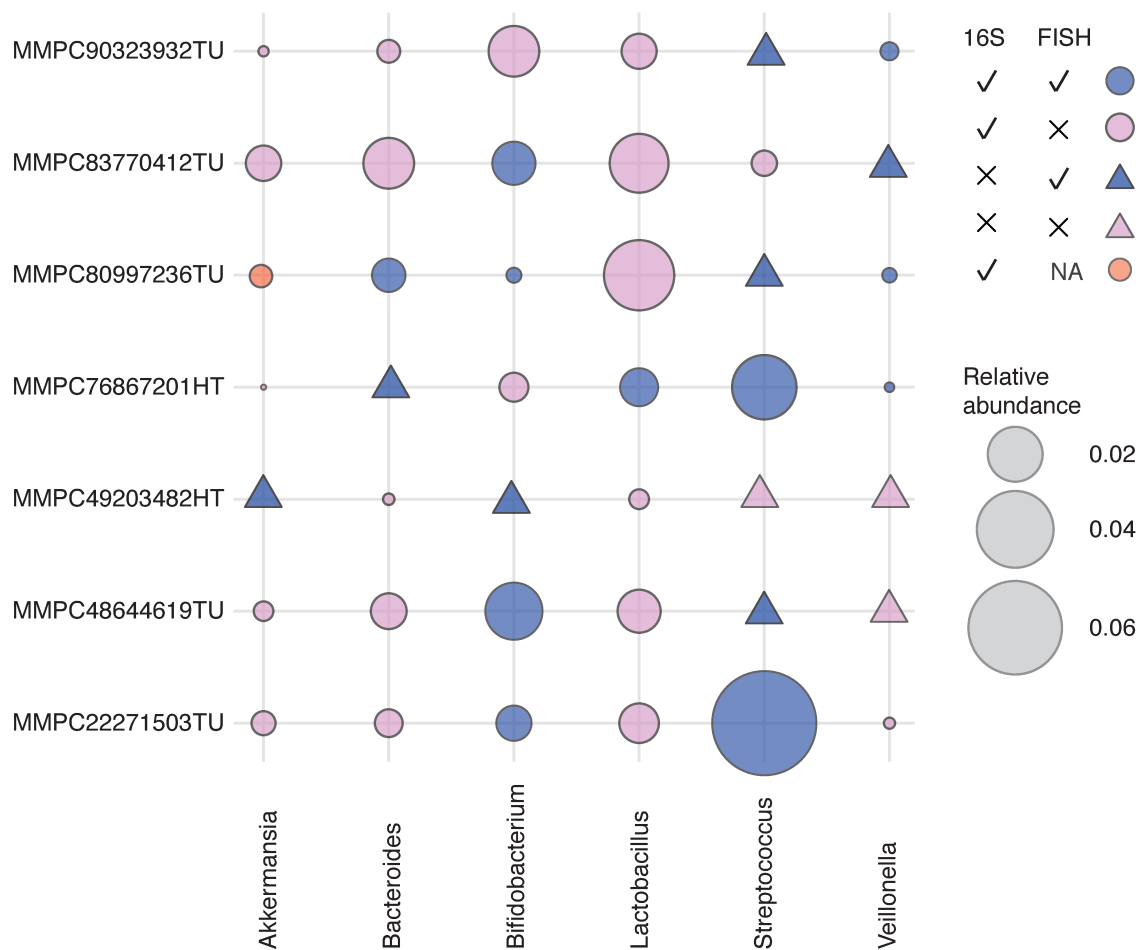


**Figure S12. The overview of external validation cohorts.** (a) Age distribution is shown for all external datasets per group. X-axis shows all the studies and y-axis displays the age distribution. (b) Sequencing depth is represented across cohorts. (c) Gender information is displayed if available as bar plot. Green is used for females while blue is for males for available studies. M:Million.



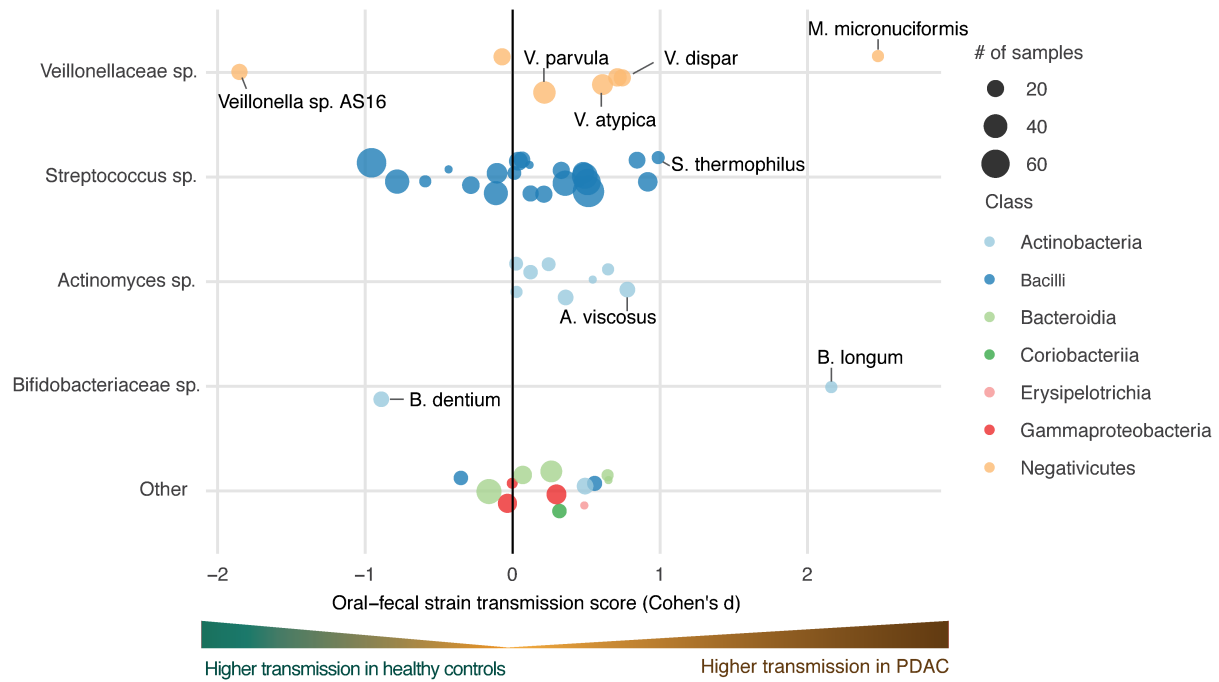
**Figure S13. Relative abundance of genera in tumor and non-tumor pancreatic tissue.**

Relative abundance of several genera is shown as bar plots. Orange is used to present the pancreatic tumor tissue, while blue is used for non-tumor tissue.



**Figure S14. Detailed information of tested samples via in-situ hybridization (FISH).**

Rows display the tested samples and columns show the tested genera. The size of the dot represents relative abundance of genus in the given sample. Triangles show that 16S was negative for given samples and color code displays if FISH was positive (blue) or negative (pink). One sample, displayed in orange, did not have enough tissue material for FISH testing. NA: Not available.



**Figure S15. Oral-fecal transmission scores differ between PDAC cases and controls.**

Oral-gut transmission scores (y-axis) of each species are displayed grouped by genus (x-axis). The number of subjects is represented by the size of the circle and the color represents the corresponding class group.

Supplementary Table S1: ES Cohort Details

sample_alias	experiment_name	experiment_type	instrument_model	replicate	insert_size	read_length	read_count	environment_material	timepoint
MMPC-3103102-ST-0	MMPC35551931ST	metagenome	Illumina HiSeq 4000	0	350	150	11417135	feces [ENVO:00002003]	0
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MMPC-3103316-ST-0	MMPC52740890ST	metagenome	Illumina HiSeq 4000	0	350	150	33343541	feces [ENVO:00002003]	0
MMPC-3103317-ST-0	MMPC48152369ST	metagenome	Illumina HiSeq 4000	0	350	150	48281105	feces [ENVO:00002003]	0
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MMPC-3103319-ST-0	MMPC11113549ST	metagenome	Illumina HiSeq 4000	0	350	150	40759382	feces [ENVO:00002003]	0
MMPC-3103319-ST-0	MMPC50507232ST	metagenome	Illumina HiSeq 4000	0	350	150	48108766	feces [ENVO:00002003]	0
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MMPC-3109109-ST-0	MMPC79962166ST	metagenome	Illumina HiSeq 4000	0	350	150	45702011	feces [ENVO:00002003]	0
MMPC-3109110-ST-0	MMPC84979047ST	metagenome	Illumina HiSeq 4000	0	350	150	51915806	feces [ENVO:00002003]	0
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MMPC-3109115-ST-0	MMPC40565253ST	metagenome	Illumina HiSeq 4000	0	350	150	24643317	feces [ENVO:00002003]	0
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MMPC-3109124-ST-0	MMPC19165288ST	metagenome	Illumina HiSeq 4000	0	350	150	30229110	feces [ENVO:00002003]	0
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MMPC-3109126-ST-0	MMPC79996713ST	metagenome	Illumina HiSeq 4000	0	350	150	45709125	feces [ENVO:00002003]	0
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MMPC-3109130-ST-0	MMPC96182596ST	metagenome	Illumina HiSeq 4000	0	350	150	76577158	feces [ENVO:00002003]	0
MMPC-3109132-ST-0	MMPC77094060ST	metagenome	Illumina HiSeq 4000	0	350	150	36697241	feces [ENVO:00002003]	0
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MMPC-3109203-ST-0	MMPC13726847ST	metagenome	Illumina HiSeq 4000	0	350	150	21663895	feces [ENVO:00002003]	0
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MMPC-3109306-ST-0	MMPC42177786ST	metagenome	Illumina HiSeq 4000	0	350	150	22749462	feces [ENVO:00002003]	0
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MMPC-3103107-SA-0	MMPC33950978OR	metagenome	Illumina HiSeq 4000	0	350	150	3047069	saliva [ENVO:02000036]	0
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MMPC-3109107-SA-0	MMPC44731247OR	metagenome	Illumina HiSeq 4000	0	350	150	8048004	saliva [ENVO:02000036]	0
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MMPC-3109305-SA-0	MMP63021713OR	165	Illumina MiSeq	0	292	292	NA	saliva [ENVO:02000036]	0
MMPC-3109306-SA-0	MMP33663665OR	165	Illumina MiSeq	0	292	292	NA	saliva [ENVO:02000036]	0
MMPC-3109315-SA-0	MMP59175809OR	165	Illumina MiSeq	0	292	292	NA	saliva [ENVO:02000036]	0
MMPC-3109319-SA-0	MMP52031211OR	165	Illumina MiSeq	0	292	292	NA	saliva [ENVO:02000036]	0
MMPC-3109324-SA-0	MMP11599193OR	165	Illumina MiSeq	0	292	292	NA	saliva [ENVO:02000036]	0

MMPC-3103102123-TU-0	MMPC52294238TU	165	Illumina MiSeq	0	292	292	16762	pancreas [UBERON:0001264]	0
MMPC-3103102125-HT-0	MMPC96437096HT	165	Illumina MiSeq	0	292	292	118983	pancreas [UBERON:0001264]	0
MMPC-3103106123-TU-0	MMPC22271503TU	165	Illumina MiSeq	0	292	292	27346	pancreas [UBERON:0001264]	0
MMPC-3103107123-TU-0	MMPC87955180TU	165	Illumina MiSeq	0	292	292	50932	pancreas [UBERON:0001264]	0
MMPC-3103108123-TU-0	MMPC54922715TU	165	Illumina MiSeq	0	292	292	31245	pancreas [UBERON:0001264]	0
MMPC-3103108125-HT-0	MMPC24466339HT	165	Illumina MiSeq	0	292	292	127835	pancreas [UBERON:0001264]	0
MMPC-3103111125-HT-0	MMPC80734101HT	165	Illumina MiSeq	0	292	292	95924	pancreas [UBERON:0001264]	0
MMPC-3103112123-TU-0	MMPC83770412TU	165	Illumina MiSeq	0	292	292	21764	pancreas [UBERON:0001264]	0
MMPC-3103112125-HT-0	MMPC40334378HT	165	Illumina MiSeq	0	292	292	91218	pancreas [UBERON:0001264]	0
MMPC-3103114123-TU-0	MMPC48644619TU	165	Illumina MiSeq	0	292	292	17849	pancreas [UBERON:0001264]	0
MMPC-3103114125-HT-0	MMPC35209818HT	165	Illumina MiSeq	0	292	292	114214	pancreas [UBERON:0001264]	0
MMPC-3103116123-TU-0	MMPC75019385TU	165	Illumina MiSeq	0	292	292	38264	pancreas [UBERON:0001264]	0
MMPC-3103116125-HT-0	MMPC89754577HT	165	Illumina MiSeq	0	292	292	84488	pancreas [UBERON:0001264]	0
MMPC-3103116125-HT-c	MMPC76929964HT	165	Illumina MiSeq	0	292	292	99306	pancreas [UBERON:0001264]	0
MMPC-3103118123-TU-0	MMPC53213093TU	165	Illumina MiSeq	0	292	292	19403	pancreas [UBERON:0001264]	0
MMPC-3103119123-TU-0	MMPC77465766TU	165	Illumina MiSeq	0	292	292	35083	pancreas [UBERON:0001264]	0
MMPC-3103119123-TU-c	MMPC24078588TU	165	Illumina MiSeq	0	292	292	71331	pancreas [UBERON:0001264]	0
MMPC-3103119125-HT-0	MMPC37714156HT	165	Illumina MiSeq	0	292	292	85195	pancreas [UBERON:0001264]	0
MMPC-3103119125-HT-c	MMPC49102349HT	165	Illumina MiSeq	0	292	292	111138	pancreas [UBERON:0001264]	0
MMPC-3103120125-HT-0	MMPC36983914HT	165	Illumina MiSeq	0	292	292	73461	pancreas [UBERON:0001264]	0
MMPC-3103121123-TU-0	MMPC90323932TU	165	Illumina MiSeq	0	292	292	32825	pancreas [UBERON:0001264]	0
MMPC-3103121123-TU-c	MMPC58485046TU	165	Illumina MiSeq	0	292	292	140836	pancreas [UBERON:0001264]	0
MMPC-3103125123-TU-0	MMPC49164313TU	165	Illumina MiSeq	0	292	292	50097	pancreas [UBERON:0001264]	0
MMPC-3103125127-HT-0	MMPC86673187HT	165	Illumina MiSeq	0	292	292	96102	pancreas [UBERON:0001264]	0
MMPC-3103129123-TU-0	MMPC80997236TU	165	Illumina MiSeq	0	292	292	64881	pancreas [UBERON:0001264]	0
MMPC-3103129125-HT-0	MMPC49203482HT	165	Illumina MiSeq	0	292	292	110366	pancreas [UBERON:0001264]	0
MMPC-3103302125-HT-0	MMPC25083626HT	165	Illumina MiSeq	0	292	292	87920	pancreas [UBERON:0001264]	0
MMPC-3103302127-HT-0	MMPC02321546HT	165	Illumina MiSeq	0	292	292	94815	ncreatic duct [UBERON:00073	0
MMPC-3103307123-TU-0	MMPC84306689TU	165	Illumina MiSeq	0	292	292	35125	pancreas [UBERON:0001264]	0
MMPC-3103307123-TU-c	MMPC63721423TU	165	Illumina MiSeq	0	292	292	101573	pancreas [UBERON:0001264]	0
MMPC-3103307125-HT-0	MMPC74146850HT	165	Illumina MiSeq	0	292	292	87273	pancreas [UBERON:0001264]	0
MMPC-3103309123-TU-0	MMPC53390762TU	165	Illumina MiSeq	0	292	292	25942	pancreas [UBERON:0001264]	0
MMPC-3103309125-HT-0	MMPC33872709HT	165	Illumina MiSeq	0	292	292	93994	pancreas [UBERON:0001264]	0
MMPC-3103330125-HT-0	MMPC35081045HT	165	Illumina MiSeq	0	292	292	75059	pancreas [UBERON:0001264]	0
MMPC-3109101123-TU-0	MMPC82234530TU	165	Illumina MiSeq	0	292	292	63990	pancreas [UBERON:0001264]	0
MMPC-3109102123-TU-0	MMPC26308927TU	165	Illumina MiSeq	0	292	292	63118	pancreas [UBERON:0001264]	0
MMPC-3109102125-HT-0	MMPC83100372HT	165	Illumina MiSeq	0	292	292	97384	pancreas [UBERON:0001264]	0
MMPC-3109103123-TU-0	MMPC79418921TU	165	Illumina MiSeq	0	292	292	92173	pancreas [UBERON:0001264]	0
MMPC-3109105123-TU-0	MMPC96387591TU	165	Illumina MiSeq	0	292	292	86922	pancreas [UBERON:0001264]	0
MMPC-3109105125-HT-0	MMPC33126922HT	165	Illumina MiSeq	0	292	292	96057	pancreas [UBERON:0001264]	0
MMPC-3109107123-TU-0	MMPC80047993TU	165	Illumina MiSeq	0	292	292	78692	pancreas [UBERON:0001264]	0
MMPC-3109107125-HT-0	MMPC65170291HT	165	Illumina MiSeq	0	292	292	66822	pancreas [UBERON:0001264]	0
MMPC-3109111123-TU-0	MMPC23067667TU	165	Illumina MiSeq	0	292	292	8	pancreas [UBERON:0001264]	0
MMPC-3109111125-HT-0	MMPC80978814HT	165	Illumina MiSeq	0	292	292	150064	pancreas [UBERON:0001264]	0
MMPC-3109111125-HT-c	MMPC42159796HT	165	Illumina MiSeq	0	292	292	116464	pancreas [UBERON:0001264]	0
MMPC-3109113123-TU-0	MMPC42025082TU	165	Illumina MiSeq	0	292	292	91603	pancreas [UBERON:0001264]	0
MMPC-3109113125-HT-0	MMPC93630826HT	165	Illumina MiSeq	0	292	292	93581	pancreas [UBERON:0001264]	0
MMPC-3109117123-TU-0	MMPC44512561TU	165	Illumina MiSeq	0	292	292	101695	pancreas [UBERON:0001264]	0
MMPC-3109117125-HT-0	MMPC58544120HT	165	Illumina MiSeq	0	292	292	76970	pancreas [UBERON:0001264]	0
MMPC-3109117127-HT-0	MMPC66546739HT	165	Illumina MiSeq	0	292	292	72952	ncreatic duct [UBERON:00073	0
MMPC-3109140123-TU-0	MMPC57986041TU	165	Illumina MiSeq	0	292	292	137586	pancreas [UBERON:0001264]	0

gender	age_years	collection_datct	disease_s	tax_id	smoker	center	alcohol_statu:	alldiab	diabcat	obese	panctype	asthma	allacid
male	79	2015-2018	PC	408170	smoker	2	0	0	0	0	0	0	0
male	62	2015-2018	PC	408170	ex-smoker	2	1	0	0	0	0	0	0
female	69	2015-2018	PC	408170	smoker	2	0	0	0	0	0	0	1
male	54	2015-2018	PC	408170	non-smoker	2	1	0	0	0	0	0	0
male	54	2015-2018	PC	408170	non-smoker	2	1	0	0	0	0	0	0
female	68	2015-2018	PC	408170	non-smoker	2	1	0	0	NA	0	0	0
male	68	2015-2018	PC	408170	ex-smoker	2	1	1	1	0	0	0	0
male	68	2015-2018	PC	408170	ex-smoker	2	1	1	1	0	0	0	0
male	71	2015-2018	PC	408170	non-smoker	2	0	0	0	0	0	0	0
male	84	2015-2018	PC	408170	non-smoker	2	1	1	1	0	0	0	0
male	84	2015-2018	PC	408170	non-smoker	2	1	1	1	0	0	0	0
female	69	2015-2018	PC	408170	non-smoker	2	0	0	0	0	0	1	1
male	66	2015-2018	PC	408170	smoker	2	1	0	0	0	0	0	1
male	82	2015-2018	PC	408170	ex-smoker	2	1	1	2	0	0	0	0
male	82	2015-2018	PC	408170	ex-smoker	2	1	1	2	0	0	0	0
male	70	2015-2018	PC	408170	non-smoker	2	1	1	2	1	0	0	0
male	70	2015-2018	PC	408170	non-smoker	2	1	1	2	1	0	0	0
male	60	2015-2018	PC	408170	smoker	2	1	0	0	0	2	0	0
male	63	2015-2018	PC	408170	ex-smoker	2	1	0	0	1	0	0	0
male	45	2015-2018	PC	408170	ex-smoker	2	1	0	0	0	NA	1	0
female	73	2015-2018	PC	408170	non-smoker	2	1	0	0	1	0	0	0
female	67	2015-2018	PC	408170	non-smoker	2	0	0	0	0	0	0	0
female	67	2015-2018	PC	408170	non-smoker	2	0	0	0	0	0	0	0
male	66	2015-2018	PC	408170	non-smoker	2	0	1	2	0	0	0	0
male	68	2015-2018	PC	408170	smoker	2	1	1	1	0	0	0	0
male	52	2015-2018	PC	408170	smoker	2	1	0	0	1	0	0	1
male	68	2015-2018	PC	408170	non-smoker	2	1	0	0	0	0	0	0
male	83	2015-2018	CTR	408170	ex-smoker	2	1	0	0	NA	0	0	0
male	57	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
male	57	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
female	70	2015-2018	CTR	408170	non-smoker	2	0	0	0	0	1	0	1
male	54	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
female	72	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	0
male	76	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	1	2	0	0
male	67	2015-2018	CTR	408170	non-smoker	2	1	1	2	0	0	0	0
male	67	2015-2018	CTR	408170	non-smoker	2	1	1	2	0	0	0	0
male	82	2015-2018	CTR	408170	non-smoker	2	0	0	0	0	0	0	0
female	64	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
male	63	2015-2018	CTR	408170	ex-smoker	2	1	0	0	0	0	0	1
male	84	2015-2018	CTR	408170	ex-smoker	2	1	1	2	1	0	0	0
male	61	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	0
male	53	2015-2018	CTR	408170	ex-smoker	2	1	0	0	0	0	0	0
male	57	2015-2018	CTR	408170	non-smoker	2	1	0	0	1	0	0	1
male	38	2015-2018	CTR	408170	ex-smoker	2	1	0	0	0	0	0	0
female	68	2015-2018	CTR	408170	ex-smoker	2	1	0	0	0	0	0	0
female	81	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	1
female	65	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
male	61	2015-2018	CTR	408170	non-smoker	2	0	0	0	0	0	0	0
male	59	2015-2018	CTR	408170	ex-smoker	2	1	0	0	1	0	0	0
male	56	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	0
male	63	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	1
male	63	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	0
male	79	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	0
male	64	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
female	62	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	1	2	1	0
male	54	2015-2018	Pancreatitis	408170	smoker	2	1	0	0	0	2	0	0
female	58	2015-2018	Pancreatitis	408170	smoker	2	1	1	1	0	2	0	1
male	58	2015-2018	Pancreatitis	408170	smoker	2	1	0	0	0	2	0	0
male	67	2015-2018	Pancreatitis	408170	smoker	2	1	0	0	0	2	0	0
male	76	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	0
female	72	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	0
male	58	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	1	2	0	0
male	78	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	0
male	60	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	1
male	58	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
male	64	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	1	1	2	0	1
male	64	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	1	1	2	0	1
male	45	2015-2018	Pancreatitis	408170	ex-smoker	2	1	0	0	0	2	0	1
female	83	2015-2018	Pancreatitis	408170	smoker	2	0	1	2	0	2	0	0
female	71	2015-2018	Pancreatitis	408170	non-smoker	2	0	0	0	0	2	1	0
male	64	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
male	61	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
male	55	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
male	62	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
female	55	2015-2018	Pancreatitis	408170	smoker	2	1	0	0	1	2	0	0
female	78	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
female	78	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
female	67	2015-2018	PC	408170	smoker	8	0	0	0	0	0	1	1
female	74	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
female	74	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
male	85	2015-2018	PC	408170	ex-smoker	8	0	0	0	0	0	0	0
female	71	2015-2018	PC	408170	non-smoker	8	0	1	1	1	0	0	1
female	68	2015-2018	PC	408170	non-smoker	8	1	1	1	0	0	0	0
female	81	2015-2018	PC	408170	non-smoker	8	1	1	1	1	0	0	0
male	86	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	0
female	71	2015-2018	PC	408170	non-smoker	8	1	0	0	0	0	0	1
male	85	2015-2018	PC	408170	ex-smoker	8	1	1	2	0	0	0	0
male	85	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	1

female	84	2015-2018	PC	408170	non-smoker	8	1	1	1	1	0	0	1
male	74	2015-2018	PC	408170	non-smoker	8	1	0	0	1	0	0	1
male	88	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	1
female	79	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	1
female	79	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	1
male	80	2015-2018	PC	408170	non-smoker	8	1	1	2	0	0	0	1
male	49	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	1	1
male	79	2015-2018	PC	408170	smoker	8	1	0	0	0	0	0	0
female	80	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
male	86	2015-2018	PC	408170	NA	8	1	0	0	1	0	0	0
male	86	2015-2018	PC	408170	NA	8	1	0	0	1	0	0	0
male	70	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	0
female	85	2015-2018	PC	408170	non-smoker	8	1	0	0	1	0	0	0
female	71	2015-2018	PC	408170	non-smoker	8	1	0	0	0	0	0	0
male	67	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	0
male	69	2015-2018	PC	408170	ex-smoker	8	1	1	2	1	0	0	0
female	75	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
female	79	2015-2018	PC	408170	non-smoker	8	0	0	0	1	0	0	0
female	74	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
male	62	2015-2018	PC	408170	ex-smoker	8	1	1	2	0	1	0	1
male	83	2015-2018	PC	408170	ex-smoker	8	1	1	2	0	0	0	0
female	65	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	0
male	56	2015-2018	PC	408170	ex-smoker	8	1	0	0	1	0	0	1
male	78	2015-2018	PC	408170	ex-smoker	8	1	0	0	1	0	0	0
male	45	2015-2018	PC	408170	smoker	8	1	0	0	1	0	0	1
male	81	2015-2018	PC	408170	non-smoker	8	1	1	2	1	0	0	0
male	64	2015-2018	PC	408170	smoker	8	1	1	1	0	0	0	0
female	88	2015-2018	CTR	408170	non-smoker	8	1	0	0	NA	0	0	0
female	80	2015-2018	CTR	408170	non-smoker	8	0	1	2	1	0	1	1
male	88	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
female	79	2015-2018	CTR	408170	non-smoker	8	1	0	0	1	0	0	0
female	59	2015-2018	CTR	408170	non-smoker	8	0	0	0	1	0	0	0
male	91	2015-2018	CTR	408170	non-smoker	8	1	0	0	0	0	0	1
female	62	2015-2018	CTR	408170	non-smoker	8	1	0	0	0	0	0	0
male	81	2015-2018	CTR	408170	ex-smoker	8	1	1	2	0	0	0	0
male	81	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	1
female	76	2015-2018	CTR	408170	non-smoker	8	0	0	0	0	0	0	0
male	71	2015-2018	CTR	408170	non-smoker	8	1	0	0	1	0	0	0
male	93	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	1	0
female	85	2015-2018	CTR	408170	smoker	8	0	0	0	0	0	0	0
male	72	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	0	0
male	49	2015-2018	CTR	408170	non-smoker	8	1	0	0	0	0	0	1
female	87	2015-2018	CTR	408170	non-smoker	8	0	1	2	1	0	0	0
male	87	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
male	78	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	0	0
male	78	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	0	0
female	88	2015-2018	CTR	408170	non-smoker	8	0	0	0	0	0	0	0
female	69	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
male	73	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
male	77	2015-2018	CTR	408170	non-smoker	8	1	0	0	0	0	0	0
female	85	2015-2018	CTR	408170	non-smoker	8	0	0	0	0	0	0	0
female	89	2015-2018	CTR	408170	non-smoker	8	1	NA	NA	0	0	1	0
male	69	2015-2018	CTR	408170	ex-smoker	8	1	0	0	1	0	0	1
female	58	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
male	59	2015-2018	CTR	408170	ex-smoker	8	0	0	0	0	0	0	0
male	71	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	1	0
female	77	2015-2018	Pancreatitis	408170	non-smoker	8	1	0	0	0	2	0	1
female	51	2015-2018	Pancreatitis	408170	ex-smoker	8	1	0	0	0	2	0	1
female	56	2015-2018	Pancreatitis	408170	non-smoker	8	1	0	0	0	2	0	1
male	74	2015-2018	Pancreatitis	408170	ex-smoker	8	1	0	0	1	2	0	0
male	54	2015-2018	Pancreatitis	408170	non-smoker	8	1	1	2	0	2	0	0
male	68	2015-2018	Pancreatitis	408170	smoker	8	0	1	2	1	2	0	0
male	79	2015-2018	PC	1679718	smoker	2	0	0	0	0	0	0	0
male	62	2015-2018	PC	1679718	ex-smoker	2	1	0	0	0	0	0	0
female	69	2015-2018	PC	1679718	smoker	2	0	0	0	0	0	0	1
male	54	2015-2018	PC	1679718	non-smoker	2	1	0	0	0	0	0	0
female	68	2015-2018	PC	1679718	non-smoker	2	1	0	0	NA	0	0	0
male	68	2015-2018	PC	1679718	ex-smoker	2	1	1	1	0	0	0	0
male	71	2015-2018	PC	1679718	non-smoker	2	0	0	0	0	0	0	0
male	84	2015-2018	PC	1679718	non-smoker	2	1	1	1	0	0	0	0
female	69	2015-2018	PC	1679718	non-smoker	2	0	0	0	0	0	1	1
male	66	2015-2018	PC	1679718	smoker	2	1	0	0	0	0	0	1
male	82	2015-2018	PC	1679718	ex-smoker	2	1	1	2	0	0	0	0
male	70	2015-2018	PC	1679718	non-smoker	2	1	1	2	1	0	0	0
male	60	2015-2018	PC	1679718	smoker	2	1	0	0	0	2	0	0
male	63	2015-2018	PC	1679718	ex-smoker	2	1	0	0	1	0	0	0
male	45	2015-2018	PC	1679718	ex-smoker	2	1	0	0	0	NA	1	0
female	73	2015-2018	PC	1679718	non-smoker	2	1	0	0	1	0	0	0
female	67	2015-2018	PC	1679718	non-smoker	2	0	0	0	0	0	0	0
male	66	2015-2018	PC	1679718	non-smoker	2	0	1	2	0	0	0	0
male	68	2015-2018	PC	1679718	smoker	2	1	1	1	0	0	0	0
male	52	2015-2018	PC	1679718	smoker	2	1	0	0	1	0	0	1
male	68	2015-2018	PC	1679718	non-smoker	2	1	0	0	0	0	0	0
male	83	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	NA	0	0	0
male	57	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	0
female	70	2015-2018	CTR	1679718	non-smoker	2	0	0	0	0	1	0	1
male	54	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	0
female	72	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	0
male	76	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	1	2	0	0
male	67	2015-2018	CTR	1679718	non-smoker	2	1	1	2	0	0	0	0

male	82	2015-2018	CTR	1679718	non-smoker	2	0	0	0	0	0	0	0
female	64	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	0
male	63	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	0	0	0	1
male	84	2015-2018	CTR	1679718	ex-smoker	2	1	1	2	1	0	0	0
male	61	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	0
male	53	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	0	0	0	0
male	57	2015-2018	CTR	1679718	non-smoker	2	1	0	0	1	0	0	1
male	38	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	0	0	0	0
female	68	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	0	0	0	0
female	81	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	1
female	65	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	0
male	61	2015-2018	CTR	1679718	non-smoker	2	0	0	0	0	0	0	0
male	59	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	1	0	0	0
male	56	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	0
male	63	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	1
male	63	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	0
male	79	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	0	2	0	0
male	64	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	0
male	54	2015-2018	Pancreatitis	1679718	smoker	2	1	0	0	0	2	0	0
male	58	2015-2018	Pancreatitis	1679718	smoker	2	1	0	0	0	2	0	0
male	58	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	1	2	0	0
male	58	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	0
male	64	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	1	1	2	0	1
female	55	2015-2018	Pancreatitis	1679718	smoker	2	1	0	0	1	2	0	0
female	78	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
female	67	2015-2018	PC	1679718	smoker	8	0	0	0	0	0	1	1
female	74	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
male	85	2015-2018	PC	1679718	ex-smoker	8	0	0	0	0	0	0	0
female	71	2015-2018	PC	1679718	non-smoker	8	0	1	1	1	0	0	1
female	68	2015-2018	PC	1679718	non-smoker	8	1	1	1	0	0	0	0
female	81	2015-2018	PC	1679718	ex-smoker	8	0	1	2	1	0	0	0
female	81	2015-2018	PC	1679718	non-smoker	8	1	1	1	1	0	0	0
male	86	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	0
female	71	2015-2018	PC	1679718	non-smoker	8	1	0	0	0	0	0	1
male	85	2015-2018	PC	1679718	ex-smoker	8	1	1	2	0	0	0	0
male	85	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	1
female	78	2015-2018	PC	1679718	non-smoker	8	0	0	0	1	0	0	0
female	84	2015-2018	PC	1679718	non-smoker	8	1	1	1	1	0	0	1
male	74	2015-2018	PC	1679718	non-smoker	8	1	0	0	1	0	0	1
male	88	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	1
female	79	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	1
male	80	2015-2018	PC	1679718	non-smoker	8	1	1	2	0	0	0	1
male	49	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	1	1
female	80	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
male	81	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
male	86	2015-2018	PC	1679718	NA	8	1	0	0	1	0	0	0
female	88	2015-2018	CTR	1679718	non-smoker	8	1	0	0	NA	0	0	0
female	68	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
female	80	2015-2018	CTR	1679718	non-smoker	8	0	1	2	1	0	1	1
male	88	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
female	79	2015-2018	CTR	1679718	non-smoker	8	1	0	0	1	0	0	0
female	59	2015-2018	CTR	1679718	non-smoker	8	0	0	0	1	0	0	0
female	77	2015-2018	CTR	1679718	non-smoker	8	1	0	0	1	0	0	0
female	84	2015-2018	CTR	1679718	non-smoker	8	0	0	0	0	0	0	0
female	84	2015-2018	CTR	1679718	non-smoker	8	0	0	0	0	0	0	0
male	91	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	1
female	62	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	0
female	62	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	0
male	81	2015-2018	CTR	1679718	ex-smoker	8	1	1	2	0	0	0	0
male	81	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	1
female	76	2015-2018	CTR	1679718	non-smoker	8	0	0	0	0	0	0	0
female	78	2015-2018	CTR	1679718	non-smoker	8	0	1	2	0	0	0	1
male	71	2015-2018	CTR	1679718	non-smoker	8	1	0	0	1	0	0	0
male	93	2015-2018	CTR	1679718	smoker	8	1	0	0	1	0	1	0
female	85	2015-2018	CTR	1679718	smoker	8	0	0	0	0	0	0	0
male	72	2015-2018	CTR	1679718	smoker	8	1	0	0	1	0	0	0
male	49	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	1
female	78	2015-2018	CTR	1679718	smoker	8	1	NA	0	0	0	0	0
female	87	2015-2018	CTR	1679718	non-smoker	8	0	1	2	1	0	0	0
male	87	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
male	87	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
male	78	2015-2018	CTR	1679718	smoker	8	1	0	0	1	0	0	0
female	77	2015-2018	Pancreatitis	1679718	non-smoker	8	1	0	0	0	2	0	1
male	74	2015-2018	Pancreatitis	1679718	ex-smoker	8	1	0	0	1	2	0	0
male	54	2015-2018	Pancreatitis	1679718	non-smoker	8	1	1	2	0	2	0	0
female	68	2015-2017	PC	646099	smoker	2	0	0	0	0	0	0	1
male	54	2015-2017	PC	646099	non-smoker	2	1	0	0	0	0	0	0
female	68	2015-2017	PC	646099	non-smoker	2	1	0	0	1	0	0	0
male	71	2015-2017	PC	646099	non-smoker	2	0	0	0	0	0	0	0
male	60	2015-2017	PC	646099	smoker	2	1	0	0	0	2	0	0
male	63	2015-2017	PC	646099	ex-smoker	2	1	0	0	1	0	0	0
male	63	2015-2017	PC	646099	ex-smoker	2	1	0	0	1	0	0	0
male	45	2015-2017	PC	646099	ex-smoker	2	1	0	0	0	0	1	0
male	45	2015-2017	PC	646099	ex-smoker	2	1	0	0	0	0	1	0
female	73	2015-2017	PC	646099	non-smoker	2	1	0	0	1	0	0	0
male	66	2015-2017	PC	646099	non-smoker	2	0	1	2	0	0	0	0
male	68	2015-2017	PC	646099	non-smoker	2	1	0	0	0	0	0	0
male	79	2015-2017	Pancreatitis	646099	ex-smoker	2	1	1	2	0	2	0	0
male	54	2015-2017	Pancreatitis	646099	NA	2	1	0	0	0	1	0	0
male	54	2015-2017	Pancreatitis	646099	NA	2	1	0	0	0	1	0	0

male	79	2015-2018	PC	408170	smoker	2	0	0	0	0	0	0	0
male	62	2015-2018	PC	408170	ex-smoker	2	1	0	0	0	0	0	0
female	69	2015-2018	PC	408170	smoker	2	0	0	0	0	0	0	1
male	54	2015-2018	PC	408170	non-smoker	2	1	0	0	0	0	0	0
female	68	2015-2018	PC	408170	non-smoker	2	1	0	0	NA	0	0	0
male	68	2015-2018	PC	408170	ex-smoker	2	1	1	1	0	0	0	0
male	71	2015-2018	PC	408170	non-smoker	2	0	0	0	0	0	0	0
male	84	2015-2018	PC	408170	non-smoker	2	1	1	1	0	0	0	0
female	69	2015-2018	PC	408170	non-smoker	2	0	0	0	0	0	1	1
male	66	2015-2018	PC	408170	smoker	2	1	0	0	0	0	0	1
male	82	2015-2018	PC	408170	ex-smoker	2	1	1	2	0	0	0	0
male	70	2015-2018	PC	408170	non-smoker	2	1	1	2	1	0	0	0
male	60	2015-2018	PC	408170	smoker	2	1	0	0	0	2	0	0
male	63	2015-2018	PC	408170	ex-smoker	2	1	0	0	1	0	0	0
male	45	2015-2018	PC	408170	ex-smoker	2	1	0	0	0	NA	1	0
female	73	2015-2018	PC	408170	non-smoker	2	1	0	0	1	0	0	0
female	67	2015-2018	PC	408170	non-smoker	2	0	0	0	0	0	0	0
male	66	2015-2018	PC	408170	non-smoker	2	0	1	2	0	0	0	0
male	68	2015-2018	PC	408170	smoker	2	1	1	1	0	0	0	0
male	52	2015-2018	PC	408170	smoker	2	1	0	0	1	0	0	1
male	68	2015-2018	PC	408170	non-smoker	2	1	0	0	0	0	0	0
male	83	2015-2018	CTR	408170	ex-smoker	2	1	0	0	NA	0	0	0
male	57	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
female	70	2015-2018	CTR	408170	non-smoker	2	0	0	0	0	1	0	1
male	54	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
female	72	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	0
male	76	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	1	2	0	0
male	67	2015-2018	CTR	408170	non-smoker	2	1	1	2	0	0	0	0
male	82	2015-2018	CTR	408170	non-smoker	2	0	0	0	0	0	0	0
female	64	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
male	63	2015-2018	CTR	408170	ex-smoker	2	1	0	0	0	0	0	1
male	84	2015-2018	CTR	408170	ex-smoker	2	1	1	2	1	0	0	0
male	61	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	0
male	53	2015-2018	CTR	408170	ex-smoker	2	1	0	0	0	0	0	0
male	57	2015-2018	CTR	408170	non-smoker	2	1	0	0	1	0	0	1
male	38	2015-2018	CTR	408170	ex-smoker	2	1	0	0	0	0	0	0
female	68	2015-2018	CTR	408170	ex-smoker	2	1	0	0	0	0	0	0
female	81	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	1
female	65	2015-2018	CTR	408170	non-smoker	2	1	0	0	0	0	0	0
male	61	2015-2018	CTR	408170	non-smoker	2	0	0	0	0	0	0	0
male	59	2015-2018	CTR	408170	ex-smoker	2	1	0	0	1	0	0	0
male	56	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	0
male	63	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	1
male	63	2015-2018	CTR	408170	smoker	2	1	0	0	0	0	0	0
male	79	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	0
male	64	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
female	62	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	1	2	1	0
male	54	2015-2018	Pancreatitis	408170	smoker	2	1	0	0	0	2	0	0
male	58	2015-2018	Pancreatitis	408170	smoker	2	1	0	0	0	2	0	0
male	67	2015-2018	Pancreatitis	408170	smoker	2	1	0	0	0	2	0	0
male	76	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	0
female	72	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	0
male	58	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	1	2	0	0
male	78	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	0
male	60	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	2	0	2	0	1
male	58	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
male	64	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	1	1	2	0	1
male	64	2015-2018	Pancreatitis	408170	ex-smoker	2	1	1	1	1	2	0	1
female	83	2015-2018	Pancreatitis	408170	smoker	2	0	1	2	0	2	0	0
female	71	2015-2018	Pancreatitis	408170	non-smoker	2	0	0	0	0	2	1	0
male	64	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
male	61	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
male	62	2015-2018	Pancreatitis	408170	smoker	2	1	1	2	0	2	0	0
female	55	2015-2018	Pancreatitis	408170	smoker	2	1	0	0	1	2	0	0
female	78	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
female	67	2015-2018	PC	408170	smoker	8	0	0	0	0	0	1	1
female	74	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
male	85	2015-2018	PC	408170	ex-smoker	8	0	0	0	0	0	0	0
female	71	2015-2018	PC	408170	non-smoker	8	0	1	1	1	0	0	1
female	68	2015-2018	PC	408170	non-smoker	8	1	1	1	0	0	0	0
female	81	2015-2018	PC	408170	non-smoker	8	1	1	1	1	0	0	0
male	86	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	0
female	71	2015-2018	PC	408170	non-smoker	8	1	0	0	0	0	0	1
male	85	2015-2018	PC	408170	ex-smoker	8	1	1	2	0	0	0	0
male	85	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	1
female	84	2015-2018	PC	408170	non-smoker	8	1	1	1	1	0	0	1
male	74	2015-2018	PC	408170	non-smoker	8	1	0	0	1	0	0	1
male	88	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	1
female	79	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	1
male	80	2015-2018	PC	408170	non-smoker	8	1	1	2	0	0	0	1
male	49	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	1	1
male	79	2015-2018	PC	408170	smoker	8	1	0	0	0	0	0	0
female	80	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
male	81	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
male	81	2015-2018	PC	408170	non-smoker	8	0	0	0	0	0	0	0
male	86	2015-2018	PC	408170	NA	8	1	0	0	1	0	0	0
male	70	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	0
female	85	2015-2018	PC	408170	non-smoker	8	1	0	0	1	0	0	0
female	71	2015-2018	PC	408170	non-smoker	8	1	0	0	0	0	0	0
female	65	2015-2018	PC	408170	ex-smoker	8	1	0	0	0	0	0	0

male	56	2015-2018	PC	408170	ex-smoker	8	1	0	0	1	0	0	1
male	78	2015-2018	PC	408170	ex-smoker	8	1	0	0	1	0	0	0
male	45	2015-2018	PC	408170	smoker	8	1	0	0	1	0	0	1
male	81	2015-2018	PC	408170	non-smoker	8	1	1	2	1	0	0	0
male	64	2015-2018	PC	408170	smoker	8	1	1	1	0	0	0	0
female	88	2015-2018	CTR	408170	non-smoker	8	1	0	0	NA	0	0	0
female	80	2015-2018	CTR	408170	non-smoker	8	0	1	2	1	0	1	1
male	88	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
female	79	2015-2018	CTR	408170	non-smoker	8	1	0	0	1	0	0	0
female	59	2015-2018	CTR	408170	non-smoker	8	0	0	0	1	0	0	0
male	91	2015-2018	CTR	408170	non-smoker	8	1	0	0	0	0	0	1
female	62	2015-2018	CTR	408170	non-smoker	8	1	0	0	0	0	0	0
male	81	2015-2018	CTR	408170	ex-smoker	8	1	1	2	0	0	0	0
male	81	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	1
female	76	2015-2018	CTR	408170	non-smoker	8	0	0	0	0	0	0	0
male	71	2015-2018	CTR	408170	non-smoker	8	1	0	0	1	0	0	0
male	93	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	1	0
female	85	2015-2018	CTR	408170	smoker	8	0	0	0	0	0	0	0
male	72	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	0	0
male	49	2015-2018	CTR	408170	non-smoker	8	1	0	0	0	0	0	1
female	87	2015-2018	CTR	408170	non-smoker	8	0	1	2	1	0	0	0
male	87	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
male	78	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	0	0
female	69	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
female	85	2015-2018	CTR	408170	non-smoker	8	0	0	0	0	0	0	0
male	69	2015-2018	CTR	408170	ex-smoker	8	1	0	0	1	0	0	1
female	58	2015-2018	CTR	408170	ex-smoker	8	1	0	0	0	0	0	0
male	59	2015-2018	CTR	408170	ex-smoker	8	0	0	0	0	0	0	0
male	71	2015-2018	CTR	408170	smoker	8	1	0	0	1	0	1	0
female	77	2015-2018	Pancreatitis	408170	non-smoker	8	1	0	0	0	2	0	1
male	74	2015-2018	Pancreatitis	408170	ex-smoker	8	1	0	0	1	2	0	0
male	54	2015-2018	Pancreatitis	408170	non-smoker	8	1	1	2	0	2	0	0
male	79	2015-2018	PC	1679718	smoker	2	0	0	0	0	0	0	0
male	62	2015-2018	PC	1679718	ex-smoker	2	1	0	0	0	0	0	0
female	69	2015-2018	PC	1679718	smoker	2	0	0	0	0	0	0	1
male	54	2015-2018	PC	1679718	non-smoker	2	1	0	0	0	0	0	0
female	68	2015-2018	PC	1679718	non-smoker	2	1	0	0	NA	0	0	0
male	68	2015-2018	PC	1679718	ex-smoker	2	1	1	1	0	0	0	0
male	71	2015-2018	PC	1679718	non-smoker	2	0	0	0	0	0	0	0
male	84	2015-2018	PC	1679718	non-smoker	2	1	1	1	0	0	0	0
female	69	2015-2018	PC	1679718	non-smoker	2	0	0	0	0	0	1	1
male	66	2015-2018	PC	1679718	smoker	2	1	0	0	0	0	0	1
male	82	2015-2018	PC	1679718	ex-smoker	2	1	1	2	0	0	0	0
male	70	2015-2018	PC	1679718	non-smoker	2	1	1	2	1	0	0	0
male	60	2015-2018	PC	1679718	smoker	2	1	0	0	0	2	0	0
male	63	2015-2018	PC	1679718	ex-smoker	2	1	0	0	1	0	0	0
male	45	2015-2018	PC	1679718	ex-smoker	2	1	0	0	0	NA	1	0
female	73	2015-2018	PC	1679718	non-smoker	2	1	0	0	1	0	0	0
female	67	2015-2018	PC	1679718	non-smoker	2	0	0	0	0	0	0	0
male	66	2015-2018	PC	1679718	non-smoker	2	0	1	2	0	0	0	0
male	68	2015-2018	PC	1679718	smoker	2	1	1	1	0	0	0	0
male	52	2015-2018	PC	1679718	smoker	2	1	0	0	1	0	0	1
male	68	2015-2018	PC	1679718	non-smoker	2	1	0	0	0	0	0	0
male	83	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	NA	0	0	0
male	57	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	0
female	70	2015-2018	CTR	1679718	non-smoker	2	0	0	0	0	1	0	1
male	54	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	0
female	72	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	0
male	76	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	1	2	0	0
male	67	2015-2018	CTR	1679718	non-smoker	2	1	1	2	0	0	0	0
male	82	2015-2018	CTR	1679718	non-smoker	2	0	0	0	0	0	0	0
female	64	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	0
male	63	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	0	0	0	1
male	84	2015-2018	CTR	1679718	ex-smoker	2	1	1	2	1	0	0	0
male	61	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	0
male	53	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	0	0	0	0
male	57	2015-2018	CTR	1679718	non-smoker	2	1	0	0	1	0	0	1
male	38	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	0	0	0	0
female	68	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	0	0	0	0
female	81	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	1
female	65	2015-2018	CTR	1679718	non-smoker	2	1	0	0	0	0	0	0
male	61	2015-2018	CTR	1679718	non-smoker	2	0	0	0	0	0	0	0
male	59	2015-2018	CTR	1679718	ex-smoker	2	1	0	0	1	0	0	0
male	56	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	0
male	63	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	1
male	63	2015-2018	CTR	1679718	smoker	2	1	0	0	0	0	0	0
male	79	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	0	2	0	0
male	64	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	0
female	62	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	1	2	1	0
male	54	2015-2018	Pancreatitis	1679718	smoker	2	1	0	0	0	2	0	0
female	58	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	1
male	58	2015-2018	Pancreatitis	1679718	smoker	2	1	0	0	0	2	0	0
male	67	2015-2018	Pancreatitis	1679718	smoker	2	1	0	0	0	2	0	0
male	76	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	0	2	0	0
female	72	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	0	2	0	0
male	58	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	1	2	0	0
male	78	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	0	2	0	0
male	60	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	2	0	1	0	1
male	58	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	0
male	64	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	1	1	1	2	0	1

male	45	2015-2018	Pancreatitis	1679718	ex-smoker	2	1	0	0	0	2	0	1
female	83	2015-2018	Pancreatitis	1679718	smoker	2	0	1	2	0	2	0	0
female	71	2015-2018	Pancreatitis	1679718	non-smoker	2	0	0	0	0	2	1	0
male	64	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	0
male	61	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	0
male	55	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	0
male	62	2015-2018	Pancreatitis	1679718	smoker	2	1	1	2	0	2	0	0
female	55	2015-2018	Pancreatitis	1679718	smoker	2	1	0	0	1	2	0	0
female	78	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
female	67	2015-2018	PC	1679718	smoker	8	0	0	0	0	0	1	1
female	74	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
male	85	2015-2018	PC	1679718	ex-smoker	8	0	0	0	0	0	0	0
female	71	2015-2018	PC	1679718	non-smoker	8	0	1	1	1	0	0	1
female	68	2015-2018	PC	1679718	non-smoker	8	1	1	1	0	0	0	0
female	81	2015-2018	PC	1679718	ex-smoker	8	0	1	2	1	0	0	0
female	81	2015-2018	PC	1679718	non-smoker	8	1	1	1	1	0	0	0
male	86	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	0
female	71	2015-2018	PC	1679718	non-smoker	8	1	0	0	0	0	0	1
male	85	2015-2018	PC	1679718	ex-smoker	8	1	1	2	0	0	0	0
male	85	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	1
female	78	2015-2018	PC	1679718	non-smoker	8	0	0	0	1	0	0	0
female	84	2015-2018	PC	1679718	non-smoker	8	1	1	1	1	0	0	1
male	74	2015-2018	PC	1679718	non-smoker	8	1	0	0	1	0	0	1
male	88	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	1
female	79	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	1
male	80	2015-2018	PC	1679718	non-smoker	8	1	1	2	0	0	0	1
male	49	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	1	1
male	79	2015-2018	PC	1679718	smoker	8	1	0	0	0	0	0	0
female	80	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
male	81	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
male	86	2015-2018	PC	1679718	NA	8	1	0	0	1	0	0	0
male	70	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	0
female	85	2015-2018	PC	1679718	non-smoker	8	1	0	0	1	0	0	0
female	71	2015-2018	PC	1679718	non-smoker	8	1	0	0	0	0	0	0
male	67	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	0
male	69	2015-2018	PC	1679718	ex-smoker	8	1	1	2	1	0	0	0
female	75	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
female	79	2015-2018	PC	1679718	non-smoker	8	0	0	0	1	0	0	0
female	74	2015-2018	PC	1679718	non-smoker	8	0	0	0	0	0	0	0
male	62	2015-2018	PC	1679718	ex-smoker	8	1	1	2	0	1	0	1
male	83	2015-2018	PC	1679718	ex-smoker	8	1	1	2	0	0	0	0
female	65	2015-2018	PC	1679718	ex-smoker	8	1	0	0	0	0	0	0
male	56	2015-2018	PC	1679718	ex-smoker	8	1	0	0	1	0	0	1
male	78	2015-2018	PC	1679718	ex-smoker	8	1	0	0	1	0	0	0
male	45	2015-2018	PC	1679718	smoker	8	1	0	0	1	0	0	1
male	81	2015-2018	PC	1679718	non-smoker	8	1	1	2	1	0	0	0
male	64	2015-2018	PC	1679718	smoker	8	1	1	1	0	0	0	0
female	88	2015-2018	CTR	1679718	non-smoker	8	1	0	0	NA	0	0	0
female	68	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
female	80	2015-2018	CTR	1679718	non-smoker	8	0	1	2	1	0	1	1
male	88	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
female	79	2015-2018	CTR	1679718	non-smoker	8	1	0	0	1	0	0	0
female	59	2015-2018	CTR	1679718	non-smoker	8	0	0	0	1	0	0	0
female	77	2015-2018	CTR	1679718	non-smoker	8	1	0	0	1	0	0	0
female	84	2015-2018	CTR	1679718	non-smoker	8	0	0	0	0	0	0	0
female	84	2015-2018	CTR	1679718	non-smoker	8	0	0	0	0	0	0	0
male	91	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	1
female	62	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	0
female	62	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	0
male	81	2015-2018	CTR	1679718	ex-smoker	8	1	1	2	0	0	0	0
male	81	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	1
female	76	2015-2018	CTR	1679718	non-smoker	8	0	0	0	0	0	0	0
female	78	2015-2018	CTR	1679718	non-smoker	8	0	1	2	0	0	0	1
male	71	2015-2018	CTR	1679718	non-smoker	8	1	0	0	1	0	0	0
male	93	2015-2018	CTR	1679718	smoker	8	1	0	0	1	0	1	0
female	85	2015-2018	CTR	1679718	smoker	8	0	0	0	0	0	0	0
male	72	2015-2018	CTR	1679718	smoker	8	1	0	0	1	0	0	0
male	49	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	1
female	78	2015-2018	CTR	1679718	smoker	8	1	NA	0	0	0	0	0
female	87	2015-2018	CTR	1679718	non-smoker	8	0	1	2	1	0	0	0
male	87	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
male	87	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
male	78	2015-2018	CTR	1679718	smoker	8	1	0	0	1	0	0	0
female	88	2015-2018	CTR	1679718	non-smoker	8	0	0	0	0	0	0	0
female	69	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
male	73	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	0
male	77	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	0	0
female	85	2015-2018	CTR	1679718	non-smoker	8	0	0	0	0	0	0	0
female	89	2015-2018	CTR	1679718	non-smoker	8	1	0	0	0	0	1	0
male	69	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	1	0	0	1
female	58	2015-2018	CTR	1679718	ex-smoker	8	1	0	0	0	0	0	1
male	59	2015-2018	CTR	1679718	ex-smoker	8	0	0	0	0	0	0	0
male	71	2015-2018	CTR	1679718	smoker	8	1	0	0	1	0	1	0
female	77	2015-2018	Pancreatitis	1679718	non-smoker	8	1	0	0	0	2	0	1
female	51	2015-2018	Pancreatitis	1679718	ex-smoker	8	1	0	0	0	2	0	1
female	51	2015-2018	Pancreatitis	1679718	ex-smoker	8	1	0	0	0	2	0	1
female	56	2015-2018	Pancreatitis	1679718	non-smoker	8	1	0	0	0	2	0	1
male	74	2015-2018	Pancreatitis	1679718	ex-smoker	8	1	0	0	1	2	0	0
male	54	2015-2018	Pancreatitis	1679718	non-smoker	8	1	1	2	0	2	0	0
male	68	2015-2018	Pancreatitis	1679718	smoker	8	0	1	2	1	2	0	0

male	79	2015-2017	PC	646099	smoker	2	0	0	0	0	0	0	0
male	79	2015-2017	PC	646099	smoker	2	0	0	0	0	0	0	0
female	68	2015-2017	PC	646099	smoker	2	0	0	0	0	0	0	1
male	54	2015-2017	PC	646099	non-smoker	2	1	0	0	0	0	0	0
female	68	2015-2017	PC	646099	non-smoker	2	1	0	0	1	0	0	0
female	68	2015-2017	PC	646099	non-smoker	2	1	0	0	1	0	0	0
male	71	2015-2017	PC	646099	non-smoker	2	0	0	0	0	0	0	0
male	84	2015-2017	PC	646099	non-smoker	2	1	1	1	0	0	0	0
male	84	2015-2017	PC	646099	non-smoker	2	1	1	1	0	0	0	0
female	69	2015-2017	PC	646099	non-smoker	2	0	0	0	0	0	1	1
female	69	2015-2017	PC	646099	non-smoker	2	0	0	0	0	0	1	1
male	82	2015-2017	PC	646099	ex-smoker	2	1	1	2	0	0	0	0
male	82	2015-2017	PC	646099	ex-smoker	2	1	1	2	0	0	0	0
male	82	2015-2017	PC	646099	ex-smoker	2	1	1	2	0	0	0	0
male	60	2015-2017	PC	646099	smoker	2	1	0	0	0	2	0	0
male	63	2015-2017	PC	646099	ex-smoker	2	1	0	0	1	0	0	0
male	63	2015-2017	PC	646099	ex-smoker	2	1	0	0	1	0	0	0
male	63	2015-2017	PC	646099	ex-smoker	2	1	0	0	1	0	0	0
male	63	2015-2017	PC	646099	ex-smoker	2	1	0	0	1	0	0	0
male	45	2015-2017	PC	646099	ex-smoker	2	1	0	0	0	0	1	0
female	73	2015-2017	PC	646099	non-smoker	2	1	0	0	1	0	0	0
female	73	2015-2017	PC	646099	non-smoker	2	1	0	0	1	0	0	0
male	66	2015-2017	PC	646099	non-smoker	2	0	1	2	0	0	0	0
male	66	2015-2017	PC	646099	non-smoker	2	0	1	2	0	0	0	0
male	68	2015-2017	PC	646099	non-smoker	2	1	0	0	0	0	0	0
male	68	2015-2017	PC	646099	non-smoker	2	1	0	0	0	0	0	0
male	79	2015-2017	Pancreatitis	646099	ex-smoker	2	1	1	2	0	2	0	0
male	79	2015-2017	Pancreatitis	646099	ex-smoker	2	1	1	2	0	2	0	0
male	54	2015-2017	Pancreatitis	646099	smoker	2	1	0	0	0	1	0	0
male	54	2015-2017	Pancreatitis	646099	smoker	2	1	0	0	0	1	0	0
male	54	2015-2017	Pancreatitis	646099	smoker	2	1	0	0	0	1	0	0
male	58	2015-2017	Pancreatitis	646099	smoker	2	1	0	0	0	0	0	0
male	58	2015-2017	Pancreatitis	646099	smoker	2	1	0	0	0	0	0	0
female	55	2015-2017	Pancreatitis	646099	smoker	2	1	0	0	1	0	0	0
female	78	2015-2017	PC	646099	non-smoker	8	0	0	0	0	0	0	0
female	67	2015-2017	PC	646099	smoker	8	0	0	0	0	0	1	1
female	67	2015-2017	PC	646099	smoker	8	0	0	0	0	0	1	1
female	74	2015-2017	PC	646099	non-smoker	8	0	0	0	0	0	0	0
female	71	2015-2017	PC	646099	non-smoker	8	0	1	1	1	0	0	1
female	71	2015-2017	PC	646099	non-smoker	8	0	1	1	1	0	0	1
female	81	2015-2017	PC	646099	ex-smoker	8	0	1	2	1	0	0	0
female	81	2015-2017	PC	646099	ex-smoker	8	0	1	2	1	0	0	0
male	85	2015-2017	PC	646099	ex-smoker	8	1	1	2	0	0	0	0
male	85	2015-2017	PC	646099	ex-smoker	8	1	1	2	0	0	0	0
male	85	2015-2017	PC	646099	ex-smoker	8	1	1	2	0	0	0	0
female	78	2015-2017	PC	646099	non-smoker	8	0	0	0	1	0	0	0
female	78	2015-2017	PC	646099	non-smoker	8	0	0	0	1	0	0	0
female	79	2015-2017	PC	646099	non-smoker	8	0	0	0	0	0	0	1
female	79	2015-2017	PC	646099	non-smoker	8	0	0	0	0	0	0	1
female	79	2015-2017	PC	646099	non-smoker	8	0	0	0	0	0	0	1
male	81	2015-2017	PC	646099	non-smoker	8	1	1	2	1	0	0	0















tetformin.ev	probiot	periodontitis	recession	FHPDAC	pack/year	inctype_patie	nasal	CA19	coded_ca19	PDAC_stage
0	0	1	NA	0	3	0	0	18	-1	2
0	0	0	0	0	1	0	0	133	1	4
0	0	0	1	0	2	0	0	126	1	2
0	0	1	1	0	0	0	1	124	1	2
0	0	1	1	0	0	0	1	124	1	2
0	0	0	0	0	0	0	0	5170	1	2
0	0	1	0	0	3	0	0	NA	0	1
0	0	1	0	0	3	0	0	NA	0	1
0	0	NA	0	0	0	0	0	0	-1	2
2	0	0	0	NA	0	0	0	55	1	2
2	0	0	0	NA	0	0	0	55	1	2
0	0	1	1	0	0	0	0	2572	1	2
0	0	1	1	0	3	0	1	151	1	3
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1	0	0	0	0	1	0	0	25	-1	2
1	0	0	0	0	0	0	0	629	1	4
1	0	0	0	0	0	0	0	629	1	4
0	0	0	0	1	3	2	0	90	1	2
0	0	0	0	0	3	0	0	326	1	2
0	0	1	1	0	2	NA	1	319	1	2
0	0	1	0	0	0	0	0	143	1	2
0	0	0	0	0	0	0	0	0	-1	3
0	0	0	0	0	0	0	0	0	-1	3
1	0	0	0	0	0	0	0	1049	1	2
0	0	1	0	0	3	0	0	2219	1	4
0	0	0	1	0	1	0	0	449	1	4
0	1	1	1	0	0	0	1	1	-1	1
0	0	NA	NA	0	3	0	0	74	1	NA
0	0	0	1	0	0	0	1	NA	0	NA
0	0	0	1	0	0	0	1	NA	0	NA
0	0	0	0	0	0	1	0	1	-1	NA
0	0	1	1	0	0	0	0	4	-1	NA
0	0	1	0	0	2	0	0	1	-1	NA
0	0	0	1	0	3	2	0	NA	0	NA
2	0	NA	0	0	0	0	0	11	-1	NA
2	0	NA	0	0	0	0	0	11	-1	NA
0	0	0	1	0	0	0	0	17	-1	NA
0	0	0	0	1	0	0	0	12	-1	NA
0	0	0	0	0	2	0	0	NA	0	NA
1	0	NA	0	0	2	0	0	NA	0	NA
0	0	0	0	0	3	0	1	NA	0	NA
0	0	0	0	0	3	0	0	8	-1	NA
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0	0	0	0	0	1	0	0	9	-1	NA
0	0	0	0	0	2	0	0	NA	0	NA
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1	0	0	1	0	0	0	0	NA	0	1
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0	0	0	1	0	0	0	0	760	1	3
1	0	0	1	1	3	0	1	19	-1	1
0	0	0	0	0	1	0	0	NA	0	1

0	0	0	0	0	0	0	0	9810	1	3
0	0	0	0	0	0	0	0	348	1	3
0	0	0	0	0	1	0	0	13617	1	4
0	0	0	1	0	0	0	0	419	1	2
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1	0	0	1	0	0	0	1	34	-1	4
0	0	0	0	0	0	0	0	351	1	3
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0	0	1	0	0	1	0	0	NA	0	2
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0	0	0	0	0	0	0	0	5	-1	NA
0	0	0	1	0	0	0	1	NA	0	NA
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0	0	0	0	0	3	0	0	15	-1	NA
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0	0	0	0	0	0	0	0	6	-1	NA
0	1	0	0	0	2	0	0	13	-1	NA
0	0	0	0	0	1	0	0	28	-1	NA
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0	0	0	1	0	0	0	0	6	-1	NA
0	0	0	0	0	0	NA	0	13	-1	NA
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0	0	0	1	0	3	NA	0	8	-1	NA
0	1	1	1	0	1	NA	0	NA	0	NA
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0	0	1	0	0	3	0	0	NA	0	1
0	0	NA	0	0	0	0	0	0	-1	2
2	0	0	0	NA	0	0	0	55	1	2
0	0	1	1	0	0	0	0	2572	1	2
0	0	1	1	0	3	0	1	151	1	3
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0	0	0	1	0	1	0	0	449	1	4
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0	0	1	0	0	2	0	0	1	-1	NA
0	0	0	1	0	3	2	0	NA	0	NA
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0	0	0	1	0	0	0	0	17	-1	NA
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0	0	0	0	0	2	0	0	NA	0	NA
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0	0	0	0	0	1	0	0	NA	0	1
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0	0	0	1	0	0	0	0	760	-1	3
1	0	0	1	1	3	0	1	19	-1	1
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1	0	0	1	0	0	0	1	34	-1	4
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0	0	0	0	0	0	0	0	25	-1	4
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0	1	0	1	0	2	0	0	7	-1	NA
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0	0	0	0	0	3	0	0	NA	0	NA
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0	0	0	0	0	0	0	0	7	-1	NA
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0	0	0	1	0	0	0	1	NA	0	NA
0	1	0	1	0	0	0	0	8	-1	NA
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0	0	0	0	0	3	0	0	15	-1	NA
0	0	0	0	0	1	0	1	NA	0	NA
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0	0	0	0	0	0	0	0	6	-1	NA
0	1	0	0	0	2	0	0	13	-1	NA
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0	0	0	0	0	1	NA	0	NA	0	NA
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0	0	0	0	0	3	0	0	11	-1	NA
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0	0	1	NA	0	3	0	0	18	-1	2
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0	0	0	1	0	2	0	0	126	1	2
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2	0	0	0	NA	0	0	0	55	1	2
0	0	1	1	0	0	0	0	2572	1	2
0	0	1	1	0	3	0	1	151	1	3
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0	0	1	1	0	2	NA	1	319	1	2
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0	0	0	0	0	0	0	0	0	-1	3
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0	0	NA	1	0	0	0	0	4862	1	4
0	0	0	0	0	1	0	0	NA	0	1
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1	0	0	0	1	0	NA	0	NA	0	3
0	0	0	1	0	3	NA	0	NA	0	4
0	0	0	0	0	0	0	0	1	-1	NA
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0	NA	0	0	0	0	0	0	NA	0	NA
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0	1	0	1	0	0	0	0	8	-1	NA
0	0	0	0	0	3	0	0	15	-1	NA
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0	1	0	0	0	2	0	0	13	-1	NA
0	0	0	0	0	1	0	0	28	-1	NA
0	0	1	0	0	3	0	1	15	-1	NA
0	0	0	0	0	0	0	0	7	-1	NA
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0	0	0	0	0	3	0	0	11	-1	NA
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0	0	0	0	0	0	NA	0	13	-1	NA
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0	0	1	NA	0	3	0	0	18	-1	2
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0	0	1	1	0	0	0	1	124	1	2
0	0	0	0	0	0	0	0	5170	1	2
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0	0	NA	0	0	0	0	0	0	-1	2
2	0	0	0	NA	0	0	0	55	1	2
0	0	1	1	0	0	0	0	2572	1	2
0	0	1	1	0	3	0	1	151	1	3
1	0	0	0	0	1	0	0	25	-1	2
1	0	0	0	0	0	0	0	629	1	4
0	0	0	0	1	3	2	0	90	1	2
0	0	0	0	0	3	0	0	326	1	2
0	0	1	1	0	2	NA	1	319	1	2
0	0	1	0	0	0	0	0	143	1	2
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1	0	0	0	0	0	0	0	1049	1	2
0	0	1	0	0	3	0	0	2219	1	4
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0	0	NA	NA	0	3	0	0	74	1	NA
0	0	0	1	0	0	0	1	NA	0	NA
0	0	0	0	0	0	1	0	1	-1	NA
0	0	1	1	0	0	0	0	4	-1	NA
0	0	1	0	0	2	0	0	1	-1	NA
0	0	0	1	0	3	2	0	NA	0	NA
2	0	NA	0	0	0	0	0	11	-1	NA
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0	0	0	0	1	0	0	0	12	-1	NA
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0	0	0	0	0	0	0	0	5	-1	NA
0	0	0	0	0	1	0	0	9	-1	NA
0	0	0	0	0	2	0	0	NA	0	NA
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0	0	0	0	0	0	0	0	15	-1	NA
0	0	0	0	0	0	0	0	6	-1	NA
0	0	0	0	1	3	0	0	NA	0	NA
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0	0	0	0	0	3	NA	0	NA	NA	NA
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0	0	0	1	0	0	0	0	30	-1	NA
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0	0	0	0	0	3	NA	0	NA	NA	NA
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1	0	0	0	0	2	NA	0	NA	NA	NA

0	0	1	1	0	3	NA	NA	NA	NA	NA
0	0	1	1	0	3	NA	NA	NA	NA	NA
0	0	0	1	0	2	NA	NA	NA	NA	NA
0	0	1	1	0	0	NA	NA	NA	NA	NA
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0	0	0	0	0	3	NA	NA	NA	NA	NA
0	0	0	0	0	3	NA	NA	NA	NA	NA
0	0	0	0	0	3	NA	NA	NA	NA	NA
0	0	1	1	0	2	NA	NA	NA	NA	NA
0	0	1	0	0	0	NA	NA	NA	NA	NA
0	0	1	0	0	0	NA	NA	NA	NA	NA
1	0	0	0	0	0	NA	NA	NA	NA	NA
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0	1	1	1	0	0	NA	NA	NA	NA	NA
1	0	0	0	0	2	NA	NA	NA	NA	NA
1	0	0	0	0	2	NA	NA	NA	NA	NA
0	0	1	0	0	3	NA	NA	NA	NA	NA
0	0	1	0	0	3	NA	NA	NA	NA	NA
0	0	1	0	0	3	NA	NA	NA	NA	NA
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0	0	0	1	0	0	NA	NA	NA	NA	NA
1	0	0	0	1	0	NA	NA	NA	NA	NA
1	0	0	0	1	0	NA	NA	NA	NA	NA

Supplementary Table S2

Variable Description	Variable Name	QES Column	Full Definition	Categories
Alcohol consumption status	alcohol_status		Ever drunk alcohol regularly, that is at least once per month for 6 months or more	NA = Missing or Don't know 0=Never drinker 1=Ever drinker
Alcohol consumption	alc_dd		Drinks/day of total alcohol in lifetime. Weighted average of the alcohol intakes at different ages, with weights equal to the total subject-specific time of investigation  Formula: $F/(age-20)$ $F = (mean(beer\_st1, beer\_s2, \dots, spirits\_st5) * (Min(age\_last, age)-20))$	<b>Continuous</b> <b>NA = Missing or Don't know</b> <b>0=Never drinker</b>
Have been diagnosed with diabetes	alldiab	G19A11, G19Alreland11	Subjects report if they have ever been diagnosed with diabetes.	NA=Missing or Don't Know 0= No 1=Yes
Have been diagnosed with diabetes categorical by time of diagnosis	diabcat	G19A11, G19B11, agec	Subjects that report having been diagnosed with diabetes are categorized by being diagnosed more or less than 2 years before the time of recruitment in the study	NA=Missing or Don't Know 1 = less than 3years 2 = more or equal to 3 years
BMI at two years prior recruitment $\geq 30$	obese	A7-1-1 and A61-1	Subjects that have BMI $\geq 30$ two years before study recruitment	NA=Missing or Don't know 0 = BMI $< 30$ 1= BMI $\geq 30$
Type of pancreatitis	panctype	G20B11	Subjects report if their type of pancreatitis if they have been diagnosed with it.  <b>è Use this variable for analyses</b>  *Pancreatitis cases forced all into the 'chronic' category*	NA=Missing or Don't know 0= No pancreatitis 1 = Acute 2 = Chronic
Type of pancreatitis	Panctype_patient	G20B11	Subjects report if their type of pancreatitis if they have been diagnosed with it. Variable exactly as reported by the patient	<b>NA=Missing or Don't know</b> <b>0= No pancreatitis</b> <b>1 = Acute</b> <b>2 = Chronic</b>
Have had asthma	asthma	G5-1-1	Subjects that report ever having asthma.	NA=Missing or Don't know 0= No 1=Yes
Have had nasal allergies (Rhinitis)	nasal	G11-1-1	Subjects that report having had nasal allergies including hay fever	NA=Missing or Don't know 0= No 1=Yes
Ever been diagnosed with heartburn	allhburn	G42A11, G42Alreland11	Subjects report if they have ever been diagnosed with Heartburn.	NA=Missing or Don't know 0= No 1=Yes
Ever been diagnosed with acid regurgitation	allacid	G42B11, G42Blreland11	Subjects report if they have ever been diagnosed with Acid regurgitation	NA=Missing or Don't know 0= No 1=Yes
Have been diagnosed with Rheumatoid arthritis	allrheum	G2311, G23Ireland11	Subjects report if they have ever been diagnosed with Rheumatoid arthritis	NA=Missing or Don't know 0= No 1=Yes
Ever been diagnosed with High blood pressure	allhbp	G4011, G40Ireland11	Subjects report if they have ever been diagnosed with High blood pressure	NA=Missing or Don't know 0= No 1=Yes
Ever been diagnosed with high blood pressure	cholesterol	G4311	Subjects report if they have ever been diagnosed with High cholesterol	NA=Missing or Don't know 0= No 1=Yes
Have had periodontitis	periodontitis	J7-1-1	Subjects that report, excluding the last two years, to been told by a dentist that they have	NA=Missing or Don't know 1=Yes

			periodontitis	0 = No
Have had receding gums	recession	J6-1-1	Subjects report having receding gums	NA=Missing or Don't know 1=Yes 0 = No
PDAC in a relative	FHPDAC	All K25 variables and all cancer location variables K26	History of PDAC (pancreatic ductal adenocarcinoma) in a relative. Subjects reported if any first degree relative had ever been diagnosed with PDAC. "Yes" was considered if any K25* variables was reported as yes and if location (any k26* variable) was reported to be PDAC. Diagnoses of other cancers, other than PDAC, in first degree relatives was considered as NO FHPDAC	NA=Missing or Don't know 0=no FHPDAC 1=yes FHPDAC
Diabetes control with diet	diabdiet	cdiabtmodifdieta	Subjects reported if their diabetes is controlled with dietary changes	NA= Missing or Don't know 0= Answered No to alldiab 1 = Yes 2 = No
Diabetes control with insulin	diabin	diabetinsulina	Subjects reported if their diabetes is controlled with insulin pump or injections	NA= Missing or Don't know 0= Answered No to alldiab 1 = Yes 2 = No
Diabetes control with medication	diabmed	diabetmedoral	Subjects reported if their diabetes is controlled with oral medication	NA= Missing or Don't know 0= Answered No to alldiab 1 = Yes 2 = No
Metformin	metformin.ever		Subjects report if ever taking metformin regularly, that is 3 months or more during the last five years	NA= Don't know 0 =No diabetes & no oral medication 1= Metformin use 2=No metformin use
Any acid regurgitation/Heartburn medication	abmedication	tratamproregur	The subject reports at least one medication of this class ( <u>regardless of their report on the disease status</u> )	NA= Missing or Don't know 0= No medication 1= Medication
Cholesterol medication	cholmedication	g37ttocolestalto	The subject reports at least one medication of this class ( <u>regardless of their report on the disease status</u> )	NA= Missing or Don't know 0= No medication 1= Medication
Aspirin/Paracetamol medication	asparmed	tomadoantiinflam	Subjects reported if they ever took aspirin or paracetamol or other anti-inflammatory medication regularly	NA = Don't know 0 = No 1 = Yes
salicylic	salicylic.ever		Subjects report if they ever took aspirin (irrespective of reportin other anti-inflammatory medication)	NA = Don't know 0 = No 1 = Yes
paracetamol	paracetamol.ever		Subjects report if they ever took paracetamol (irrespective of reportin other anti-inflammatory medication)	NA = Don't know 0 = No 1 = Yes
Corticosteroids medication	cortmed	H3-1-1tocorticoides	Subjects reported if they ever took corticoids regularly	NA = Missing or Don't know 0 = No 1 = Yes
NSAIDs medication	nsaidmed	H2-1-1	Subjects reported if they ever took nsaid regularly  *Answer from Sweden was based on variable H2B_1EU, if nsaid reported then a number 1 was given for this variable	NA = missing or Don't know 0 = No 1 = Yes
Antibiotics	antibiotic	h3attoantibiot	Subjects reported if they ever took antibiotics during the past five years	NA = Don't know 0 = No 1 = Yes
Pro/prebiotics	probiot	h4attoprobiotprebiot	Subjects reported if they ever took <u>prebiotics or probiotics</u> regularly during the past five years	NA = Don't know 0 = No 1 = Yes

Supplementary Table S3: DE Cohort Details

sample_alias	instrument_model	replicate	insert_size	read_length	read_count	ronment_mat	timepoint	gender	age_years	ollection_datct	disease_s
MBJT10257157ST	Illumina HiSeq 4000	0	300	150	47393312	[ENVO:0000;	0	female	65	2014-2020	PC
MBJT13029530ST	Illumina HiSeq 4000	0	300	150	44500100	[ENVO:0000;	0	male	34	2014-2020	CTR
MBJT13966914ST	Illumina HiSeq 4000	0	300	150	55422086	[ENVO:0000;	0	male	66	2014-2020	PC
MBJT14741058ST	Illumina HiSeq 4000	0	300	150	54925804	[ENVO:0000;	0	male	29	2014-2020	CTR
MBJT18793872ST	Illumina HiSeq 4000	0	300	150	48908442	[ENVO:0000;	0	female	66	2014-2020	PC
MBJT19172978ST	Illumina HiSeq 4000	0	300	150	36491564	[ENVO:0000;	0	female	54	2014-2020	CTR
MBJT19739988ST	Illumina HiSeq 4000	0	300	150	43913396	[ENVO:0000;	0	male	59	2014-2020	PC
MBJT22022613ST	Illumina HiSeq 4000	0	300	150	37301894	[ENVO:0000;	0	male	61	2014-2020	CTR
MBJT22320456ST	Illumina HiSeq 4000	0	300	150	39863688	[ENVO:0000;	0	male	74	2014-2020	PC
MBJT23133014ST	Illumina HiSeq 4000	0	300	150	44285414	[ENVO:0000;	0	female	40	2014-2020	CTR
MBJT24075712ST	Illumina HiSeq 4000	0	300	150	38908806	[ENVO:0000;	0	male	65	2014-2020	PC
MBJT24417068ST	Illumina HiSeq 4000	0	300	150	46462326	[ENVO:0000;	0	male	49	2014-2020	CTR
MBJT24770375ST	Illumina HiSeq 4000	0	300	150	41886800	[ENVO:0000;	0	female	73	2014-2020	PC
MBJT25397146ST	Illumina HiSeq 4000	0	300	150	38837880	[ENVO:0000;	0	female	68	2014-2020	CTR
MBJT26681707ST	Illumina HiSeq 4000	0	300	150	39728784	[ENVO:0000;	0	female	57	2014-2020	PC
MBJT28436807ST	Illumina HiSeq 4000	0	300	150	43156950	[ENVO:0000;	0	male	63	2014-2020	PC
MBJT28723736ST	Illumina HiSeq 4000	0	300	150	34666406	[ENVO:0000;	0	female	79	2014-2020	PC
MBJT30331144ST	Illumina HiSeq 4000	0	300	150	31449568	[ENVO:0000;	0	female	76	2014-2020	PC
MBJT32951253ST	Illumina HiSeq 4000	0	300	150	42555236	[ENVO:0000;	0	female	78	2014-2020	PC
MBJT34036667ST	Illumina HiSeq 4000	0	300	150	41733736	[ENVO:0000;	0	male	55	2014-2020	CTR
MBJT34220088ST	Illumina HiSeq 4000	0	300	150	48977086	[ENVO:0000;	0	male	80	2014-2020	CTR
MBJT35001732ST	Illumina HiSeq 4000	0	300	150	45479874	[ENVO:0000;	0	female	40	2014-2020	PC
MBJT35016775ST	Illumina HiSeq 4000	0	300	150	47763580	[ENVO:0000;	0	female	81	2014-2020	PC
MBJT35058600ST	Illumina HiSeq 4000	0	300	150	51795444	[ENVO:0000;	0	female	71	2014-2020	PC
MBJT40306752ST	Illumina HiSeq 4000	0	300	150	46477424	[ENVO:0000;	0	female	24	2014-2020	CTR
MBJT40435704ST	Illumina HiSeq 4000	0	300	150	45919556	[ENVO:0000;	0	male	63	2014-2020	PC
MBJT40701160ST	Illumina HiSeq 4000	0	300	150	50045710	[ENVO:0000;	0	female	26	2014-2020	CTR
MBJT40707177ST	Illumina HiSeq 4000	0	300	150	40242120	[ENVO:0000;	0	female	82	2014-2020	PC
MBJT41036209ST	Illumina HiSeq 4000	0	300	150	42245120	[ENVO:0000;	0	male	67	2014-2020	PC
MBJT41854002ST	Illumina HiSeq 4000	0	300	150	40271702	[ENVO:0000;	0	male	46	2014-2020	CTR
MBJT42733650ST	Illumina HiSeq 4000	0	300	150	35444906	[ENVO:0000;	0	male	80	2014-2020	PC
MBJT42917972ST	Illumina HiSeq 4000	0	300	150	40122088	[ENVO:0000;	0	female	72	2014-2020	CTR
MBJT44556531ST	Illumina HiSeq 4000	0	300	150	53434320	[ENVO:0000;	0	female	50	2014-2020	PC
MBJT46290466ST	Illumina HiSeq 4000	0	300	150	37640290	[ENVO:0000;	0	female	49	2014-2020	CTR
MBJT47281689ST	Illumina HiSeq 4000	0	300	150	55830262	[ENVO:0000;	0	female	76	2014-2020	PC
MBJT47376024ST	Illumina HiSeq 4000	0	300	150	38754652	[ENVO:0000;	0	female	66	2014-2020	PC
MBJT47759212ST	Illumina HiSeq 4000	0	300	150	31813116	[ENVO:0000;	0	male	70	2014-2020	PC
MBJT47974019ST	Illumina HiSeq 4000	0	300	150	38234074	[ENVO:0000;	0	female	81	2014-2020	PC
MBJT54042978ST	Illumina HiSeq 4000	0	300	150	51374756	[ENVO:0000;	0	female	50	2014-2020	CTR
MBJT54598736ST	Illumina HiSeq 4000	0	300	150	46583366	[ENVO:0000;	0	male	28	2014-2020	CTR
MBJT54773511ST	Illumina HiSeq 4000	0	300	150	45819402	[ENVO:0000;	0	male	31	2014-2020	CTR
MBJT55041082ST	Illumina HiSeq 4000	0	300	150	25828972	[ENVO:0000;	0	male	80	2014-2020	PC
MBJT55763017ST	Illumina HiSeq 4000	0	300	150	37164202	[ENVO:0000;	0	female	73	2014-2020	PC
MBJT56486370ST	Illumina HiSeq 4000	0	300	150	60275548	[ENVO:0000;	0	male	87	2014-2020	CTR
MBJT57869938ST	Illumina HiSeq 4000	0	300	150	36431696	[ENVO:0000;	0	female	61	2014-2020	PC
MBJT58818096ST	Illumina HiSeq 4000	0	300	150	42617446	[ENVO:0000;	0	female	69	2014-2020	PC
MBJT59111766ST	Illumina HiSeq 4000	0	300	150	39629386	[ENVO:0000;	0	female	55	2014-2020	CTR
MBJT61614909ST	Illumina HiSeq 4000	0	300	150	32278494	[ENVO:0000;	0	female	56	2014-2020	PC
MBJT61989978ST	Illumina HiSeq 4000	0	300	150	44899094	[ENVO:0000;	0	male	67	2014-2020	CTR
MBJT63497322ST	Illumina HiSeq 4000	0	300	150	41550048	[ENVO:0000;	0	female	65	2014-2020	PC
MBJT64178099ST	Illumina HiSeq 4000	0	300	150	45792268	[ENVO:0000;	0	female	28	2014-2020	CTR
MBJT64323881ST	Illumina HiSeq 4000	0	300	150	51879638	[ENVO:0000;	0	female	87	2014-2020	PC
MBJT65314632ST	Illumina HiSeq 4000	0	300	150	40100360	[ENVO:0000;	0	male	77	2014-2020	PC
MBJT66677231ST	Illumina HiSeq 4000	0	300	150	38271172	[ENVO:0000;	0	male	59	2014-2020	CTR
MBJT69789353ST	Illumina HiSeq 4000	0	300	150	37463630	[ENVO:0000;	0	female	79	2014-2020	PC
MBJT71206045ST	Illumina HiSeq 4000	0	300	150	37446348	[ENVO:0000;	0	male	64	2014-2020	PC
MBJT73212715ST	Illumina HiSeq 4000	0	300	150	29746146	[ENVO:0000;	0	male	81	2014-2020	PC
MBJT73621698ST	Illumina HiSeq 4000	0	300	150	46013560	[ENVO:0000;	0	male	22	2014-2020	CTR
MBJT75354344ST	Illumina HiSeq 4000	0	300	150	42836122	[ENVO:0000;	0	male	84	2014-2020	PC
MBJT75856433ST	Illumina HiSeq 4000	0	300	150	38763576	[ENVO:0000;	0	female	60	2014-2020	PC
MBJT76001019ST	Illumina HiSeq 4000	0	300	150	41432142	[ENVO:0000;	0	female	36	2014-2020	CTR
MBJT77767153ST	Illumina HiSeq 4000	0	300	150	51551804	[ENVO:0000;	0	female	70	2014-2020	CTR
MBJT79123647ST	Illumina HiSeq 4000	0	300	150	40277574	[ENVO:0000;	0	female	72	2014-2020	PC
MBJT82870648ST	Illumina HiSeq 4000	0	300	150	47315674	[ENVO:0000;	0	male	30	2014-2020	CTR
MBJT83133832ST	Illumina HiSeq 4000	0	300	150	40995200	[ENVO:0000;	0	male	67	2014-2020	PC
MBJT83294902ST	Illumina HiSeq 4000	0	300	150	37680998	[ENVO:0000;	0	male	64	2014-2020	CTR
MBJT85450199ST	Illumina HiSeq 4000	0	300	150	47792460	[ENVO:0000;	0	male	76	2014-2020	PC
MBJT85948313ST	Illumina HiSeq 4000	0	300	150	44611384	[ENVO:0000;	0	male	49	2014-2020	CTR
MBJT87587928ST	Illumina HiSeq 4000	0	300	150	43175984	[ENVO:0000;	0	female	62	2014-2020	CTR
MBJT89319187ST	Illumina HiSeq 4000	0	300	150	43302384	[ENVO:0000;	0	female	40	2014-2020	PC
MBJT90536787ST	Illumina HiSeq 4000	0	300	150	44711476	[ENVO:0000;	0	female	64	2014-2020	CTR
MBJT92561737ST	Illumina HiSeq 4000	0	300	150	40739728	[ENVO:0000;	0	female	32	2014-2020	CTR
MBJT92636570ST	Illumina HiSeq 4000	0	300	150	36248620	[ENVO:0000;	0	male	57	2014-2020	PC
MBJT93071877ST	Illumina HiSeq 4000	0	300	150	45583612	[ENVO:0000;	0	female	56	2014-2020	CTR
MBJT94530819ST	Illumina HiSeq 4000	0	300	150	44454590	[ENVO:0000;	0	female	84	2014-2020	PC
MBJT97792954ST	Illumina HiSeq 4000	0	300	150	42824760	[ENVO:0000;	0	male	68	2014-2020	PC

bmi	smoker	medication	stage	stage_detail	metastasis (he)	Treatment	iy_of_operati	erapy_after_G	of_last_follo	date_of_follo	Ca_19_9
27	smoker	NA	2	2b	0	1	27.02.20	1	0	May 20	1702
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
23,8	non-smoker	NA	2	2b	0	1	21.02.20	1	0	02.06.20	2
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
29,7	non-smoker	NA	2	2a	0	4	31.01.20	3	0	03.06.20	1172
NA	NA	L-Thyrox	NA	NA	NA	NA	NA	NA	NA	NA	NA
21,47	non-smoker	NA	4	4	0	6	NA	3	NA	NA	1066
27,78	smoker	NA	NA	NA	0	1	01.04.19	NA	NA	NA	1575
26,01	non-smoker	NA	1	1b	0	1	08.01.19	2	0	19.06.20	17081
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
29,54	smoker	NA	2	2b	0	1	04.07.19	2	0	06.03.20	2
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
28,2	smoker	NA	4	4	NA	8	NA	0	0	16.09.20	4
20,2	non-smoker	NA	NA	NA	0	7	09.07.19	2	0	08.05.20	1412
39,68	smoker	NA	3	3	0	4	08.01.19	3	1	08.11.19	1000
30,37	smoker	NA	3	3	0	2	26.01.19	2	0	15.06.20	71091
22,1	non-smoker	NA	4	4	0	1	04.11.19	1	0	10.08.20	5097
32,8	non-smoker	NA	4	4	0	1	10.02.20	0	0	23.04.20	2183
22,68	non-smoker	NA	4	4	1	4	06.05.19	4	1	17.06.20	2
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
32,3	non-smoker	NA	2	2a	0	1	20.08.19	2	0	14.07.20	164583
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
19,7	smoker	NA	3	3	0	1	03.03.20	1	0	30.03.20	2346
29,76	smoker	NA	4	4	1	4	04.02.19	3	1	17.03.20	3447
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
27,1	non-smoker	NA	2	2b	1	3	NA	2, 3	1	04.05.20	379
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
24,22	smoker	NA	4	4	0	8	NA	NA	0	16.09.20	138
23,26	non-smoker	NA	4	4	0	1	30.07.19	0	0	10.09.20	906
NA	smoker	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
26,8	non-smoker	NA	2	2b	0	3	16.04.19	0	1	13.07.19	160969
31,24	smoker	NA	NA	NA	0	1	17.04.19	NA	NA	NA	834
18,73	non-smoker	NA	3	3	0	1, 7	30.09.19	5	0	unknown	2
NA	NA	L-Thyrox	NA	NA	NA	NA	NA	NA	NA	NA	NA
22,7	non-smoker	NA	3	3	0	2	04.02.20	2	0	04.06.20	6021
22,39	non-smoker	NA	4	4	0	5	03.06.19	0	1	03.07.19	118805
27,13	non-smoker	NA	1	1a	0	1, 3	1.2016-01.07.	0	0	19.07.20	4
19,38	smoker	NA	3	3	NA	1	23.01.20	3	NA	16.09.20	985
28,9	non-smoker	NA	NA	NA	0	4	14.11.19	NA	NA	NA	76814
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
33,03	non-smoker	NA	2	2a	0	3	02.08.19	2	1	Jan 20	611609
25,64	non-smoker	NA	4	4	1	4	30.01.19	3	1	09.04.19	42
21,5	non-smoker	NA	NA	NA	1	2	18.02.20	2	0	11.09.20	239
32,87	non-smoker	NA	2	2a	0	1	12.04.19	2	0	20.05.20	19635
25,35	non-smoker	NA	3	3	0	1	14.02.19	2	1	24.01.20	130723
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
23,92	smoker	NA	2	2b	0	1	20.03.19	2	0	08.06.20	27812
27,47	non-smoker	NA	NA	NA	0	1	13.02.19	NA	NA	NA	16658
30,4	smoker	NA	1	1b	0	3	10.07.19	NA	NA	NA	222
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
24,14	non-smoker	NA	2	2b	0	1	29.01.19	0	1	30.01.19	7009
28,73	non-smoker	NA	2	2a	0	2	24.01.19	0	1	29.03.19	7212
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
27,8	non-smoker	NA	4	4	0	1	20.12.19	2	0	06.08.20	87632
NA	smoker	NA	NA	NA	0	NA	NA	4	NA	NA	12779
26,9	non-smoker	NA	3	3	0	3	20.02.20	NA	NA	NA	3464
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
28,04	non-smoker	NA	1	1b	NA	8	NA	0	0	16.09.20	379
25,9	non-smoker	NA	4	4	1	4	16.01.19	0	1	07.03.19	978
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
25,83	non-smoker	NA	NA	NA	0	2	13.03.19	NA	NA	NA	1865
29,76	non-smoker	NA	3	3	0	3	02.05.19	2	0	12.05.20	2498
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
22,58	non-smoker	NA	3	3	1	7	01.10.14	2	0	16.09.20	451
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
24,61	non-smoker	NA	3	3	0	3	18.07.19	NA	NA	NA	55435
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
26,29	non-smoker	NA	NA	NA	0	6	NA	NA	0	18.03.20	380838
24,22	smoker	NA	3	3	0	2	21.02.19	2	0	04.05.20	140631
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
NA	NA	no	NA	NA	NA	NA	NA	NA	NA	NA	NA
22,9	non-smoker	NA	3	3	0	1	19.12.19	2	0	Aug 20	1146
NA	NA	L-Thyrox	NA	NA	NA	NA	NA	NA	NA	NA	NA
19,92	non-smoker	NA	3	3	1	1	06.09.18	2	1	21.08.19	NA
23,84	non-smoker	NA	2	2a	NA	8	NA	3	1	.04.2020 and	142

alcohol	alcohol_	alcohol_	antibiotics	PPI	recent_medications	other_drugs	dialysis	ca19_9	histological	staging	time
1	3-4 per week	1	0	1	NA	1	0	1	N1L0V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	NA	1	0	1	NA	1	0	-1	N0L0V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	1 per week	1	0	1	NA	1	0	1	N0L0V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	-	1	0	0	7	2	0	1	IV	single sample	
1	daily	1	0	1	NA	1	0	-1	NA	ay before operation	
1	ex abus	1	1	0	NA	1	1	1	N0L0V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	NA	1	0	0	NA	1	1	-1	N0V0Pn1	G3Ray before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	-	1	1	1	1, 3, 4, 5, 7	1, 4	0	-1	IV	single sample	
1	3-4 per week	1	0	0	NA	1	0	1	NA	ay before operation	
0	-	1	0	1	2, 3, 4, 5, 7	1	0	1	III	single sample	
1	daily	1	0	0	NA	0	1	1	N2L0V0Pn1	Gay before operation	
1	1-2 per week	1	1	1	NA	1	0	1	NA	ay before operation	
0	NA	1	0	1	NA	1	0	-1	I1(hep)L0V0P	ay before operation	
1	1-2 per week	1	0	0	NA	3	0	-1	NA	ay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	NA	1	1	1	NA	1	0	1	3G2; pT3bN0	Lay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	1 per week	1	0	0	NA	1	0	1	N2L0V1Pn1	Gay before operation	
0	NA	1	1	1	NA	2	0	-1	NA	ay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	occasionally	1	0	1	6, 7	1	0	1	pN1 (2/18), L	Csingle sample	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	-	1	1	1	NA	1	0	1	IV	single sample	
1	daily	1	1	1	NA	1	0	-1	I1(lym)L0V0P	ay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	NA	1	0	0	NA	1	0	1	N1L0V0Pn1	Gay before operation	
1	daily	0	0	0	NA	0	0	-1	NA	ay before operation	
1	consumption sir	1	0	1	2, 5, 6, 7	1	1	-1	III	single sample	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	NA	1	0	0	NA	1	0	1	N2L0V0Pn1	Gay before operation	
0	NA	1	0	0	NA	1, 4	1	1	NA	adjuvant radiochemotherapy	
0	-	1	0	1	2, 4, 5, 7	6	1	-1	IA	single sample	
0	-	1	1	0	2, 6, 2007	6	0	1	III	single sample	
0	NA	1	0	0	NA	1, 3	1	1	NA	ay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	NA	1	1	1	NA	1	1	1	N0L0V0Pn1	Gay before operation	
0	NA	1	0	1	NA	1	1	1	NA	ay before operation	
0	NA	1	0	0	NA	1	0	-1	NA	ay before operation	
0	NA	1	0	1	NA	1	1	1	N0L0V0Pn1	Gay before operation	
0	NA	0	0	0	NA	0	0	1	N2L0V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	daily	1	0	0	NA	1	0	1	N1L0V0Pn1	Gay before operation	
1	daily	1	1	1	NA	1	1	1	NA	ay before operation	
0	NA	1	0	0	NA	1	0	-1	N0L0V0Pn0	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	1 per week	1	0	0	NA	1	0	1	N1L0V0Pn1	Gay before operation	
0	NA	1	0	1	NA	1	0	1	N0L0V1Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	NA	1	0	0	NA	1	0	1	NA	ay before operation	
1	daily	1	0	1	NA	3	0	1	NA	adjuvant chemotherapy	
0	NA	1	0	0	NA	1	0	-1	N2L1V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	-	1	1	0	1, 2, 3, 5, 7	1	1	1	Ib	single sample	
1	1 per week	0	1	0	NA	0	0	-1	NA	ay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	1 per month	1	0	0	NA	2	0	-1	NA	ay before operation	
0	NA	1	0	0	NA	1	0	1	N1L0V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	alcohol since 2	1	0	1	2, 5, 2007	5	1	1	1 (1/12), L1,	Vsingle sample	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	1 per month	1	0	0	NA	1	0	1	N2L1V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	NA	1	0	1	NA	1	0	1	NA	ay before operation	
0	NA	1	0	0	NA	5	0	1	N2L0V1Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
1	daily	1	1	0	NA	1	0	1	N2L1V0Pn1	Gay before operation	
NA	NA	NA	NA	NA	NA	NA	NA	0	NA	NA	
0	-	1	0	1	1, 7	1, 6	0	-1	III	single sample	
1	lay in the last	1	0	1	1, 7	1	1	1	IIA	single sample	

## Supplementary Table S4

Meta variable	Stool			Saliva		
	Sum sq	p value	adj p	Sum sq	p value	adj p
abmedication	7,40E+08	0.434882370	1.0	7,26E+07	0.726039062	1.0
Age	5,57E+10	0.348999552	1.0	1,43E+10	0.992133250	1.0
Alcohol consumption	3,28E+09	0.105017752	1.0	4,22E+07	0.796587916	1.0
Acid regurgitation	1,92E+09	0.215874045	1.0	2,05E+07	0.857472013	1.0
Diabetes	1,47E+07	0.914256854	1.0	4,78E+08	0.387311299	1.0
Heart burn	7,20E+08	0.445550072	1.0	2,11E+09	0.059764110	1.0
High blood pressure	8,65E+06	0.934099012	1.0	4,70E+06	0.931832163	1.0
Rheumatoid arthritis	7,36E+08	0.444113445	1.0	3,28E+07	0.821293457	1.0
Antibiotic	6,18E+07	0.825159237	1.0	3,55E+08	0.455050912	1.0
Aspirin/paracetamol	1,36E+09	0.297843618	1.0	1,12E+09	0.181428572	1.0
Asthma	1,76E+09	0.236597169	1.0	4,57E+08	0.395506158	1.0
Center	4,90E+09	0.046953480	1.0	2,76E+06	0.947458361	1.0
Cholesterol	1,72E+09	0.246154381	1.0	2,93E+08	0.493014836	1.0
Cholmedication	6,33E+05	0.982226159	1.0	1,48E+08	0.663216095	1.0
Corticosteroids	2,24E+09	0.179684582	1.0	4,11E+08	0.406201881	1.0
cpy1	7,16E+10	0.825078663	1.0	2,39E+10	0.800141666	1.0
Bilirubin direct	1,32E+11	0.543089087	1.0	5,08E+10	0.857634747	1.0
Bilirubin lab	1,30E+11	0.455465155	1.0	4,85E+10	0.802784617	1.0
FHPDAC	2,40E+06	0.964420558	1.0	4,78E+08	0.383616587	1.0
Gender	1,04E+09	0.362627665	1.0	5,98E+08	0.330808453	1.0
Jaundice	9,45E+09	0.004406831	0.3437328	4,30E+08	0.412200163	1.0
Jaundice imputed	8,32E+09	0.009192609	0.7078309	4,41E+08	0.403644362	1.0
Library_size	8,15E+09	0.086373435	1.0	1,55E+09	0.487294778	1.0
Metformin usage	3,98E+09	0.205930855	1.0	3,12E+08	0.783170829	1.0
Obesity	8,37E+08	0.415414037	1.0	3,49E+08	0.450252695	1.0
Paracetamol	4,49E+09	0.058802839	1.0	2,03E+09	0.072918539	1.0
Periodontitis	1,73E+08	0.710712567	1.0	1,31E+09	0.136666489	1.0
Probiotic	1,11E+07	0.925379621	1.0	1,09E+07	0.894062783	1.0
Receding gums	8,17E+08	0.415025012	1.0	2,69E+08	0.508708572	1.0
Salicylic acid	1,85E+09	0.227061461	1.0	1,94E+08	0.582723500	1.0
Salicylic acid	1,85E+09	0.227061461	1.0	1,94E+08	0.582723500	1.0
Smoking	9,62E+07	0.782721018	1.0	2,24E+09	0.059311754	1.0
Direct Bilirubin	2,75E+09	0.138132461	1.0	1,71E+09	0.102796407	1.0
Total Bilirubin	7,13E+08	0.440177205	1.0	2,87E+09	0.031822359	1.0

**Supplementary Table S5**

Meta variable	Stool		Saliva	
	R2 Bray-Curtis	p value	R2 Bray-Curtis	p value
status	0.023120267	0.0001	0.010468749	0.6507
center	0.011138008	0.2069	0.016751291	0.1285
age	0.013765355	0.0320	0.018667727	0.0680
gender	0.010546251	0.2884	0.017940759	0.0884
jaundice_imp	0.015980165	0.0087	0.007371362	0.9560
diabetes	0.013610607	0.0396	0.008990599	0.8264
obesity	0.008734297	0.6915	0.016455787	0.1449
smoking	0.008943945	0.6466	0.015490596	0.1926
alcohol_con	0.009730450	0.4545	0.017867998	0.0929
periodontitis	0.009018863	0.6237	0.009449706	0.7709
cholesterol	0.007848892	0.8750	0.010637989	0.6368
metformin	0.008315828	0.7739	0.018288711	0.0898
salicylic	0.009192529	0.5749	0.008768629	0.8462
antibiotic	0.007858670	0.8685	0.006825246	0.9740
aspirin/paracetamol	0.012544261	0.0847	0.020375695	0.0432
corticosteroids	0.010196054	0.3492	0.017140805	0.1277
asthma	0.008577892	0.7244	0.014670641	0.2412
acid regurgitation	0.011797158	0.1335	0.006447749	0.9801
rheumatoid arthritis	0.008798014	0.6747	0.011646596	0.5123
probiotic	0.009681840	0.4578	0.011088990	0.5759
paracetamol	0.010198223	0.3573	0.009887242	0.7242
heartburn	0.008810470	0.6760	0.013602585	0.3241
high blood pressure	0.007920655	0.8614	0.010496978	0.6491
receding gums	0.008765518	0.6873	0.007806693	0.9229
FHPDAC	0.010506425	0.2887	0.012944276	0.3752
acid med	0.010032433	0.3870	0.007517951	0.9390

Supplementary Table S6

Study	Diagnosis	Cohort siz	Pred.pos	Pred.neg	FPR	Model	Accession number
Average BRCA	BRCA	62			0,35	Model 1	
Average BRCA	BRCA	62			0,05	Model 2	
Average CD	CD	340			0,35	Model 1	
Average CD	CD	340			0,05	Model 2	
Average CRC	CRC	679			0,25	Model 1	
Average CRC	CRC	679			0,04	Model 2	
Average CTR	CTR	3872			0,17	Model 1	
Average CTR	CTR	3872			0,01	Model 2	
Average LD	LD	237			0,26	Model 1	
Average LD	LD	237			0,07	Model 2	
Average T1D	T1D	87			0,29	Model 1	
Average T1D	T1D	87			0,00	Model 2	
Average T2D	T2D	382			0,23	Model 1	
Average T2D	T2D	382			0,02	Model 2	
Average UC	UC	352			0,18	Model 1	
Average UC	UC	352			0,01	Model 2	
Buschart 2016	CTR	26	5	21	0,19	Model 1	
Buschart 2016	T1D	27	8	19	0,30	Model 1	PRJNA289586
Buschart 2016	CTR	26	0	26	0,00	Model 2	
Buschart 2016	T1D	27	0	27	0,00	Model 2	
Dhakan 2019	CTR	110	19	91	0,17	Model 1	PRJNA397112
Dhakan 2019	CTR	110	2	108	0,02	Model 2	
Feng 2015	CTR	63	7	56	0,11	Model 1	
Feng 2015	CRC	93	11	82	0,12	Model 1	PRJEB7774
Feng 2015	CTR	63	2	61	0,03	Model 2	
Feng 2015	CRC	93	2	91	0,02	Model 2	
FijiCOMP	CTR	172	14	158	0,08	Model 1	PRJNA217052
FijiCOMP	CTR	172	1	171	0,01	Model 2	
FMT Vaughn	CD	15	4	11	0,27	Model 1	PRJNA321058
FMT Vaughn	CD	15	1	14	0,07	Model 2	
Franzosa 2018	CTR	56	5	51	0,09	Model 1	
Franzosa 2018	UC	76	10	66	0,13	Model 1	
Franzosa 2018	CD	88	19	69	0,22	Model 1	PRJNA400072
Franzosa 2018	CTR	56	0	56	0,00	Model 2	
Franzosa 2018	UC	76	0	76	0,00	Model 2	
Franzosa 2018	CD	88	2	86	0,02	Model 2	
He 2017	CTR	54	11	43	0,20	Model 1	
He 2017	CD	49	33	16	0,67	Model 1	PRJEB15371
He 2017	CTR	54	0	54	0,00	Model 2	
He 2017	CD	49	5	44	0,10	Model 2	
Price-Lloyd 2019	UC	84	26	58	0,31	Model 1	
Price-Lloyd 2019	CD	151	38	113	0,25	Model 1	
Price-Lloyd 2019	CTR	65	5	60	0,08	Model 1	PRJNA398089
Price-Lloyd 2019	UC	84	1	83	0,01	Model 2	
Price-Lloyd 2019	CD	151	2	149	0,01	Model 2	
Price-Lloyd 2019	CTR	65	0	65	0,00	Model 2	

Hoyles 2018	LD	73	3	70	0,04	Model 1	PRJEB14215
Hoyles 2018	LD	73	3	70	0,04	Model 2	
Karlsson 2013	CTR	43	8	35	0,19	Model 1	PRJEB1786
Karlsson 2013	T2D	102	14	88	0,14	Model 1	
Karlsson 2013	CTR	43	0	43	0,00	Model 2	
Karlsson 2013	T2D	102	0	102	0,00	Model 2	
Kuang 2019	CTR	59	18	41	0,31	Model 1	PRJEB18755
Kuang 2019	T1D	29	9	20	0,31	Model 1	
Kuang 2019	CTR	59	0	59	0,00	Model 2	
Kuang 2019	T1D	29	0	29	0,00	Model 2	
Liu 2016	CTR	110	5	105	0,05	Model 1	PRJNA328899
Liu 2016	CTR	110	0	110	0,00	Model 2	
Mardinoglu 2018	LD	48	4	44	0,08	Model 1	PRJNA420817
Mardinoglu 2018	LD	48	0	48	0,00	Model 2	
Forslund 2015	CTR	372	85	287	0,23	Model 1	224, PRJEB1220, PRJ
Forslund 2015	T2D	78	16	62	0,21	Model 1	
Forslund 2015	T1D	31	8	23	0,26	Model 1	
Forslund 2015	UC	192	20	172	0,10	Model 1	
Forslund 2015	CD	37	13	24	0,35	Model 1	
Forslund 2015	CTR	372	0	372	0,00	Model 2	
Forslund 2015	T2D	78	0	78	0,00	Model 2	
Forslund 2015	T1D	31	0	31	0,00	Model 2	
Forslund 2015	UC	192	1	191	0,01	Model 2	
Forslund 2015	CD	37	1	36	0,03	Model 2	
Qin 2012	CTR	185	51	134	0,28	Model 1	PRJNA422434
Qin 2012	T2D	183	76	107	0,42	Model 1	
Qin 2012	CTR	185	1	184	0,01	Model 2	
Qin 2012	T2D	183	2	181	0,01	Model 2	
Qin 2014	LD	116	77	39	0,66	Model 1	PRJEB6337
Qin 2014	CTR	114	25	89	0,22	Model 1	
Qin 2014	LD	116	20	96	0,17	Model 2	
Qin 2014	CTR	114	1	113	0,01	Model 2	PRJNA299502
Sankaranarayanan 2015	T2D	19	3	16	0,16	Model 1	
Sankaranarayanan 2015	T2D	19	1	18	0,05	Model 2	PRJNA319574
Schirmer 2016	CTR	471	14	457	0,03	Model 1	
Schirmer 2016	CTR	471	0	471	0,00	Model 2	
Spanish	CP	29	4	25	0,14	Model 1	
Spanish	CP	29	5	24	0,17	Model 2	PRJEB12449
Vogtmann 2016	CRC	51	13	38	0,25	Model 1	
Vogtmann 2016	CTR	52	11	41	0,21	Model 1	
Vogtmann 2016	CRC	51	0	51	0,00	Model 2	
Vogtmann 2016	CTR	52	2	50	0,04	Model 2	ERP005534 PRJEB27
Wirbel 2019	CRC	60	12	48	0,20	Model 1	
Wirbel 2019	CTR	60	4	56	0,07	Model 1	
Wirbel 2019	CRC	60	1	59	0,02	Model 2	
Wirbel 2019	CTR	60	0	60	0,00	Model 2	PRJEB9576
Xie 2016	CTR	250	35	215	0,14	Model 1	
Xie 2016	CTR	250	2	248	0,01	Model 2	

Yachida 2019	CRC	352	88	264	0,25	Model 1	
Yachida 2019	CTR	289	76	213	0,26	Model 1	
Yachida 2019	CRC	352	28	324	0,08	Model 2	PRJDB4176
Yachida 2019	CTR	289	8	281	0,03	Model 2	
Yassour 2018	CTR	42	3	39	0,07	Model 1	
Yassour 2018	CTR	42	0	42	0,00	Model 2	PRJNA290381
Yu 2017	CRC	74	25	49	0,34	Model 1	
Yu 2017	CTR	54	14	40	0,26	Model 1	
Yu 2017	CRC	74	2	72	0,03	Model 2	PRJEB10878
Yu 2017	CTR	54	0	54	0,00	Model 2	
Zeevi 2015	CTR	900	81	819	0,09	Model 1	
Zeevi 2015	CTR	900	1	899	0,00	Model 2	PRJEB11532
Zeller 2014	CTR	83	10	73	0,12	Model 1	
Zeller 2014	CRC	49	18	31	0,37	Model 1	
Zeller 2014	CTR	83	4	79	0,05	Model 2	PRJEB6070, PRJEB26
Zeller 2014	CRC	49	5	44	0,10	Model 2	
Zhu 2018	BRCA	62	22	40	0,35	Model 1	
Zhu 2018	CTR	71	25	46	0,35	Model 1	
Zhu 2018	BRCA	62	3	59	0,05	Model 2	PRJNA453965
Zhu 2018	CTR	71	0	71	0,00	Model 2	

**Supplementary Table S7**

Target species/genus	Sequence	Probe	Dye	Reference
Bifidobacterium (genus)	5'- GATAGGACGCGACCCCAT -3'	Bif228	Cy3	<a href="http://probase.csb.univie.ac.at/pb_report/probe/3959">http://probase.csb.univie.ac.at/pb_report/probe/3959</a>
Veillonella (genus)	5'- AGACGCAATCCCCTCCTT -3'	Veil223	FITC	<a href="http://probase.csb.univie.ac.at/pb_report/probe/553">http://probase.csb.univie.ac.at/pb_report/probe/553</a>
Akkermansia (genus/species)	5'- CCTTGCGGTTGGCTTCAGAT -3'	MUC-1437	FITC	<a href="http://probase.csb.univie.ac.at/pb_report/probe/3898">http://probase.csb.univie.ac.at/pb_report/probe/3898</a>
Lactobacillus (genus)	5'- ACATGGAGTTCCACT -3'	Lact663	FITC	<a href="https://repositorium.sdum.uminho.pt/bitstream/1822/24334/1/pp.pdf">https://repositorium.sdum.uminho.pt/bitstream/1822/24334/1/pp.pdf</a>
Bacteroides (genus)	5'- CCAATGTGGGGGACCTT -3'	Bac303	Cy3	<a href="https://onlinelibrary.wiley.com/doi/pdf/10.1111/j.1365-2672.2011.05039.x">https://onlinelibrary.wiley.com/doi/pdf/10.1111/j.1365-2672.2011.05039.x</a>
Streptococcus (genus)	5'-TTTAGCCGTCCCTTTCTGG -3'	Strc493	Cy3	<a href="http://probase.csb.univie.ac.at/pb_report/probe/964">http://probase.csb.univie.ac.at/pb_report/probe/964</a>