**Multi-cohort analysis of Enteric fungal microbiota dysbiosis and ecological alterations in CRC**

## Introduction

### Background introduction of CRC

World Health Organization (WHO) estimated that cancer ranks the first or second in the leading causes of death before 70 years in 112 countries and ranks the third or fourth in a further 23 countries among 183 countries1. Worldwide, there were an estimated 19.3 million new cancer cases (18.1 million excluding nonmelanoma skin cancer) and 10.0 million cancer deaths (9.9 million excluding nonmelanoma skin cancer) in 20202. Colorectal cancer (CRC) is the third most commonly diagnosed malignancy and the fourth most deadly cancer, with almost 900,000 deaths annually worldwide. Its burden is expected to increase by 60% to more than 2.2 million new cases and 1.1 million cancer deaths by 20303,4.

### Fungi profile in mammal host

LATER

### Relationship between gut bacterial and fungal microbiome alterations and host diseases

LATER

## Results

### Consistent pre-procession and filtering of published and new data for the meta-analysis of CRC metagenomes.

In this meta-analysis, nine cohorts from 8 published studies 5–12 were included. The data contain fecal shotgun metagenomic sequences used to characterize the microbiome alteration of CRC patients compared to healthy individuals or adenoma patients. The 9th indoor cohort was included sequencing data from individuals with all three different clinical situations (healthy people, adenoma sufferers, and CRC patients). We performed shotgun metagenomic sequencing of stool microbiome from 417 cases with normal adenoma and CRC recruited in Hong Kong. Therefore, 2,052 samples were included, which were collected from geographically diverse metagenomic studies from 8 countries and four continents. The median of Bacteria and Eukaryota abundance are 107.19 and 104.31, respectively. The ratio of Eukaryota and Bacteria is 10-2.80, which is consistent with previous research that revealed that eukaryotes, especially fungi, consist of approximately 0.1% gut microbes13 [fig. 1a]. The exclusion criteria included three main sessions, sample sequencing quality control, suspected contamination samples filtering, and low-fungal sequencing depth selection [fig. 1b]. After excluding 723 subjects, 454 and 350, respectively, well-characterized patients with CRC and adenoma, and 525 no-CRC health controls were enrolled in this study [tab. 1]. Notably, the fecal specimens of the North American study performed the 12 cycles of limited-cycle PCR were discarded due to differences with universal metagenomics sequencing library building.

We only considered 1,329 cases for the downstream analysis (table 1) (see method). Notably, the fecal specimens of the North American study were performed the 12 cycles of limited-cycle PCR

## Discussion

### 3.1

## Methods

### 4.1

## Data availability