**Protocol Title**

*Indicate the goal of the procedure and include the model system or organism*

Author 11, #, Author 21, 2, #, $, Author 32 and Author 42, \*

*Indicate First name, Middle name initial, and Last name*

1Dept/Center, Institution name, City, Country; 2Dept/Center, Institution name, City, Country; $Current/Present address: Dept/Center, Institution name, City, Country

\*For correspondence: email

#Contributed equally to this work

**[Abstract]** Enter text here.

*Introduce the research field (1-2 sentences), provide context by describingmentioning the*      *existing methodologies*     *(1-2 sentences), summarize the approach/protocol (3-4 sentences max), and finish by summarizing the advantages of the approach/protocol being presented (1-2 sentences).*

**Graphic abstract:** (optional)

Insert the figure here.

*It could be a flowchart of the method/procedures, the highlight of the key steps or the innovative design from this protocol etc.*

1. *Resolution: a minimum of 500 dpi.*
2. *Minimize the usage of text in the picture*

**Keywords:** Keyword 1, Keyword 2, Keyword 3 …

*5-8 keywords to help searches.*

**[Background]**

*Briefly, introduce the research field that*      *the software/pipeline can be used*     *. Mention*      *related methodologies and summarize the advantages of using your approach*     *.*      *If possible, elaborate on other possible applications of the software/pipeline*      *. (****Reference citation format: “Dow et al., 2018”****).*

**Software**

1. cutadapt (Martin, 2011; version 2.10; https://cutadapt.readthedocs.io/en/stable/changes.html#v2-1-2019-03-15)
2. ITSxpress (Rivers et al., 2018; version 1.0; https://github.com/usda-ars-gbru/itsxpress)
3. R (R Core Team, 2017; version 3.6.3; https://www.R-project.org/)
4. tidyverse (Wickham et al., 2019; version 1.3.0; https://www.tidyverse.org/)
5. DADA2 (Callahan et al., 2016; version 1.14.1; https://benjjneb.github.io/dada2/)
7. Software 1 name (Citation; Version; Website)
8. Software 2 name (Citation; Version; Website)
9. …

*List individual software separately, especially in cases where a long software workflow is used.*

**Input data**

* + - 1. Description of the format of input data
      2. Explanation of example data that should be either included in the supplementary materials or generated through provided codes

**Case study**

*Step-by-step guide for the utility of the software/pipeline*

* + - 1. …
      2. …

1. ***Write with active voice and verbs in the present tense*** *(e.g., write "Parse the output files from the homology searches” instead of “The output files from the homology searches were parsed.").*
2. ***Provide representative data (intermediary and final) and notes/tips to help others:*** *Your protocol should provide enough information for a first-year graduate student to perform it and be successful.*
3. ***Codes and Scripts:*** *the codes or scripts could be presented in the Case study section or submitted as supplementary file(s).*
4. ***For images:***
5. *Resolution: a minimum of 300 dpi.*
6. *Text in Figure: 8-12 point.*
7. *Panel label: “A, B, C…” (uppercase, bold). Use the same typeface for all figures.*
8. *Each figure should have a title and a comprehensive but concise legend.*
9. *Include scale bars where it applies (e.g. microscope images).*
10. *Figures should be embedded in the text.*
11. ***For tables:***
12. *Each table should have a brief title and a comprehensive but concise legend.* *Put table title above the table. Consistently use “****Table 1. Title.*** *Xxx.”, indicate Table 1. 2... in the text.*
13. *Each table should be editable. Prepare all tables using the table function in a word-processing program.*
14. *Each table should have the following three horizontal lines:*

*One under the title, above the column heading, 1 pound;*

*One between the column headings and the body of the table, 0.75 pounds;*

*One at the bottom of the table, 1 pound.*

1. *Each table should be set off from the text close to where it is first cited.*
2. ***For videos:*** *typical smart phone camera videos are usually adequate*
   * 1. *Each video should have a title.*
     2. *Each video file should be less than 1 GB.*
     3. *Each video must be submitted as a separate file.*

**Result interpretation**

*Interpretation/insight upon the data analysis above*

**Discussion (optional)**

*Justification or suggestion of selection of methods and/or key parameters; perspectives of future method development.*

**Notes** (optional)

*Provide general comments such as notes about reproducibility and variability in your hands and cautionary points.*

**Acknowledgments**

*Include the following information: 1) acknowledge funding sources; 2) acknowledge previous work or the original research paper where this protocol was derived from.*

**Competing interests**

1. *The corresponding author should provide a statement of financial and non-financial competing interests on behalf of all authors.*
2. *Examples include paid employment or consultancy, stock ownership, patent applications, personal relationships with individuals involved in the submission or evaluation of a protocol, and receipt of funding or free products from the vendors of the reagents/equipment or other advertisers.*

**Ethics**

1. *All protocols that have used human and/or animal subjects must mention the specific ethics committee that approved the described experiment.*
2. *Protocols including human subjects should also indicate that informed consent was obtained from all subjects.*

**References**

Dow, L. E., O'Rourke, K. P., Simon, J., Tschaharganeh, D. F., van Es, J. H., Clevers, H. and Lowe, S. W. (2015). Apc restoration promotes cellular differentiation and reestablishes crypt homeostasis in colorectal cancer. *Cell* 161(7): 1539-1552.

*Strongly recommended: providing the references in our standard format (see above) and listing them in the order of aliphatic.*