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Protocol status: Working We use this protocol and it's working

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SOP for DSS kinetics studies

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ABSTRACT

SOP for DSS kinetics studies

MATERIALS

- Autoclaved tap water
- Dextran sulfate sodium salt (MW = 40,000, ThermoFisher #J63606-22)
- Sterilized drinking bottles
- Hemoccult SENSA Fecal Occult Blood Slide Test System (Fisher #SK-64152)
- o This should include applicator sticks, single slides, and slide developer droppers
 - Daily log(s) for animal weights and DAI
- Small boxes to weigh and house animals temporarily until stools are passed we use empty pipette tip boxes



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BEFORE STARTING:

1 Ensure you have garnered the appropriate approval from your institution's regulatory body over animal use in research to conduct experiments with DSS, and comply with any additional instructions they may provide to ensure your research animals are being properly cared for.

In cages with multiple animals, ensure animals have unique identifiers. We routinely notch ears to distinguish animals and further put unique marks on tails with a red marker to more easily and quickly distinguish animals.

PROTOCOL:

- Weigh each mouse from a cage individually in small boxes. Record their weights in grams to the nearest tenth for that day.
- 3 Keep each mouse in their own box until stools are passed. For a healthy mouse, this should occur quickly. A mouse can be scruffed to expedite this if needed.
- Once stool is passed, return the mouse to its home cage. While doing this, inspect the mouse's rectum and look for bleeding and/or smeared stool. These features reflect more severe colitis and will be used in DAI scoring later.
- With an applicator stick, pick up the stool and smear it onto one of the designated areas of a slide. Observe the appearance and overall feel of the stool as it spreads onto the slide. Also, observe whether blood is

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visible in the smear.

- **6** Open the other side of the slide and observe whether there is any soaking through from the side to which the stool was applied.
- 7 Score the stool consistency according to the rubric below:

| | A | В | |
|---|---|--|--|
| | 0 Dry and compact, crumbles instead of smears | | |
| | 1 | Slightly loose and "gooey" | |
| 2 Same as 1 plus there is observable soakage on the back of | | Same as 1 plus there is observable soakage on the back of the card | |
| | 3 N/A | | |
| | 4 | Same as 2 plus there is stool smeared to the mouse's rectum | |

- 8 Add two drops of developer solution to the backside of the card and to the quality control panel below.
- 9 After at least 30s, verify that the positive control dot is blue and the negative control dot is not.
- 10 Look back to the side to which the stool was applied. If you can see blue, consider the sample as positive.
- 11 Then, score the fecal blood according to the rubric below:

| A | В | |
|---|--|--|
| 0 | No blood in stool and negative Hemoccult reading | |
| 1 | N/A | |
| 2 | No blood in stool but positive Hemoccult reading | |
| 3 | Blood in stool and positive Hemoccult reading | |

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| А | В |
|---|---|
| 4 | Same as 3 but blood is also visible at the mouse's rectum |

12 Score the weight loss according to the rubric below:

| A | В |
|---|---------------------------|
| 0 | Less than 2% weight loss |
| 1 | 2.0 - 7.99% weight loss |
| 2 | 8.0 - 13.99% weight loss |
| 3 | 14.0 - 19.99% weight loss |
| 4 | Over 20% weight loss |

13 For statistical analysis and data visualization, sum all three scores into one total DAI score.