



Frequently Asked Questions

Amylose/Amylopectin Assay Kit

Q. 1: With your amylose/amylopectin kit can you calculate the amylose content from the Con A supernatant absorbance, without performing the total starch section if you have recorded the weight of the sample?

A: Amylose is always reported as a percentage of the starch, not as a percentage of the sample weight. The method should be followed as described.

Q. 2: I would like to know if your Amylose/Amylopectin kit will also give the total starch content?

A: Yes, it is possible to get total starch content as well as amylose/amylopectin ratio using the Amylose kit. However, we recommend the Total Starch Assay Kit as the method of choice for this.

Q. 3: In your Amylose/Amylopectin Kit can you please tell me what modifications for steps A2 – A7 can be used for problem samples which do not appear to dissolve fully in DMSO (e.g. rice flours, potato starch)?

A: I suggest that you make the following changes:

1. A2 – use 2 ml DMSO
2. A4 – use total of 12 ml Ethanol (4 + 8 – need longer tubes)
3. A6 – use 2 ml of DMSO
4. A7 – use 4 ml of Con A solvent.

Of these, the changes in A6 and A7 are probably the most important.

Q. 4: What is the difference between your Amylose/Amylopectin kit and the Total Starch Kit? I need to separate amylose from amylopectin in maize plants and quantify each fraction and wonder which kit would be most suitable.

A: The Amylose/Amylopectin kit employs Con A to selectively precipitate the amylopectin.

Q. 5: Do you provide any information in your Amylose/Amylopectin kit for the separation of the two starch fractions?

A: I think that the only effective method to separate amylose and amylopectin is by gel filtration.

Q. 6: We are using your Amylose/Amylopectin Assay Kit. When we measure the Amylose/Amylopectin content of Sagopalm and rice starch (powder form), white precipitate are remaining and not dissolved completely though we add DMSO and heat a tube. Also, when we add DMSO after the precipitation by adding ethanol, white precipitate is remaining. What is the reason for this? Can you kindly let us know a counter measure?

A: The white precipitate is most probably protein, and this will cause no real problems. However, an alternative is as follows:

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Step 2: Dissolve in 2 ml DMSO, Centrifuge at high speed (could use a micro-centrifuge – 14,000 rpm, but bench centrifuge – 3,000 rpm, may be OK) and recover the supernatant. Treat this as follows:

Step 4: Add 4 ml ethanol, then 8 ml of ethanol.

Step 6: Use 2 ml of DMSO

Step 7: Use 4 ml of Con A solvent and then make up to 25 ml as per method.

Q. 7: We are analysing amylose using your kit K-AMYL. Do you have any recommendations regarding sample grinding? We have used a Retsch laboratory mill with a 0.5mm screen. The particle size may be important for starch extraction.

A: Milling with Retsch 0.5 mm screen is what we do.

Q. 8: I am interested in your Amylose/Amylopectin Assay Kit. Is this assay suitable for potato starch?

A: The method should be fine for amylose/amylopectin in potato starch.

Q. 9: Could you please give us details about the reproducibility of the Amylose/Amylopectin kit?

A: Reproducibility +/- 2 to 4% of the value.

Q. 10: I have a question regarding the amylose/amylopectin kit. In the ConA precipitation step, after the hour incubation, is the centrifugation speed or temperature more important, or are they equally important? I don't have a microfuge capable of that speed that also cools. I could put the microfuge in a refrigerated room (temperature approximately 5°C).

A: Speed of centrifugation is the most important parameter. We use the microfuge at room temperature.

Q. 11: How sensitive is your Amylose/Amylopectin kit – can it differentiate between 50/50 and a 55/45 ratio?

A: Yes, there should be no problem in distinguishing samples with these differences in amylose content.

Q. 12: I have trouble with Steps A4 and A6 of your Amylose/Amylopectin Kit – i.e. distributing or dissolving the sample in DMSO without forming gelatinous lumps (starch) or grimy material that sticks to the sides of the tube (flour). How can I avoid this or cope with it?

A: Rice starch should dissolve completely, but the solution may not be "crystal clear". In fact, it generally isn't. With rice flour, the solution will be quite turbid due to materials other than starch, which will not dissolve. You can also increase the amount of DMSO used in the test. We have now introduced this modification into the method.

Q. 13: I have a question regarding your amylose/amylopectin assay kit. The solution (supernatant) is not always clear after centrifuging (A5) and after mixing with Con A solvent (A7) – depending on the sample. Is this a problem or is this OK?

A: Solution not always clear A5 – this is ok.
Solution not always clear A7 – this is ok as well.

Q. 14: What kind of flour is the reference starch sample in your Amylose/Amylopectin kit from (potato, rice, wheat)?

A: The starch control is high amylose maize starch.