

## Result Interpretation

- Ensure QC criteria are met.
- The concentration of Dextran is determined by comparing the sample absorbance to the absorbance of the positive control:
  - Samples with a lower absorbance (less color) than the 300 ppm control have a concentration of Dextran greater than the concentration of the control.
  - Samples with an absorbance that falls between the 0 and the 300 ppm controls have a concentration of Dextran greater than 0 and less than 300 ppm.

Based on a statistically significant number of tests, the 300 ppm control's acceptable percent binding ratio (%B/B<sub>0</sub>), determined as two standard deviations from the mean, ranges between 38.7 – 45.3%. From this, the 300 ppm control must fall within this specified %B/B<sub>0</sub> range for actionable, semi-quantitative results.

%B/B<sub>0</sub> (%Bound/Maximum Bound): Ratio of the absorbance (OD) of a particular sample or standard tube to that of the maximum binding (B<sub>0</sub>) well, (OD of 0 ppm control).

## Technical Assistance

For questions regarding this kit or for additional information about Beacon products, contact us.

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## Safety

Stop Solution is 1N hydrochloric acid. Handle with care. To receive complete safety information on this product, contact Beacon Analytical Systems, Inc., and request Safety Data Sheets.

## General Limited Warranty

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## Intended Use

The Beacon Dextran 40 Tube Kit is an immunoassay for the detection of Dextran in fresh beet juice samples. This product is intended for research use only.

## Principles

Dextran HRP Enzyme Conjugate is pipetted into the test tubes followed by the Controls and the Sample Extract(s). A soluble polyclonal Dextran antibody solution is then added to the test tubes to initiate the reaction. During an incubation, Dextran in the control/sample and Dextran HRP Enzyme Conjugate compete for binding sites to the Dextran Antibody in solution. The Dextran antibodies are then bound by a second Sheep  $\alpha$ -Rabbit antibody immobilized on the test tubes. Following an incubation, the tubes are washed to remove any unbound Dextran and Dextran HRP Enzyme Conjugate. After washing, a colorless substrate is added to the tubes and any bound enzyme conjugate will convert the substrate to a blue color. Following an incubation, the reaction is stopped with the addition of Stop Solution and the amount of color in each tube is measured. The color of the unknown sample is compared to the color of the controls and the Dextran concentration of the sample is derived.

## Reagents and Materials Provided

- 2 Units Bags each containing 20 test tubes, vacuum sealed in aluminized pouch with desiccant.
- 2 X 7 mL Vials of Dextran Controls (0 and 300 ppm). Dextran T- 40 Calibrators from Sigma Aldrich.  
**Note:** The 300 ppm control contains 1/100th of the stated value to account for the 1:100 dilution during sample preparation. No further correction is required to obtain the concentration of Dextran in the sample.
- 1 X 12 mL Bottle of Dextran HRP Enzyme Conjugate.
- 1 X 12 mL Bottle of Dextran Antibody.
- 1 X 25 mL Bottle of Substrate.
- 1 X 25 mL Bottle of Stop Solution.
- 2 Units Packets of Wash Concentrate (dilute prior to use).

## Reagents and Materials Required but Not Provided

- Pipette(s) with disposable tips capable of dispensing the required volume(s).
- Micro-Centrifuge tubes and Micro-Centrifuge capable of required rpm(s).
- Laboratory quality distilled or deionized water.
- Reagents and materials for sample preparation.
- Personal protective equipment.
- Paper towels or equivalent absorbent material.
- Wash bottle.
- Tube rack.
- Timer.
- Photometer capable of reading absorbance at 450 nm in 12 mm x 75 mm tubes.
- Repeater pipette(s) with disposable tips capable of dispensing the required volume(s) (recommended if running more than five tubes at once).

### Kit Handling Notes and Precautions

- Read the product brochure in its entirety prior to use.
- The kit, in its original packaging, can be used until the end of the month indicated on the box label.
- Do not use reagents after their expiration date.
- Store all kit components at 2°C to 8°C (36°F to 46°F) when not in use.
- Reagents should be brought to room temperature, 20°C to 28°C (68°F to 82°F), prior to use. Avoid prolonged (> 24 hours) storage at room temperature.
- Do not freeze kit components or expose them to temperatures greater than 37°C (99°F).
- Running Controls and Samples in duplicate will improve assay precision and accuracy.
- Precise transfer of samples and reagents by using a calibrated pipette that is capable of dispensing the required volume is critical to obtain proper assay results.
- If running more than five tubes at once, the use of a repeater pipette is recommended when adding the Dextran Antibody, Substrate and Stop Solution.
- All procedural steps should be completed without interruption. Ensure all reagents, materials and equipment are ready at the appropriate time.
- Each reagent is optimized for use in the Beacon Dextran 40 Tube Kit. Do not substitute reagents from any other manufacturer into the test kit. Do not combine reagents from other Beacon Dextran 40 Tube Kits with different lot numbers.
- Do not reuse test tubes.
- Dilution or adulteration of reagents or samples not called for in the procedure may result in inaccurate results.
- Damage to or obstruction of the optical surface may cause unsatisfactory results.

### Specificity

The Beacon Dextran Tube Kit can detect several Dextran molecular weights. The percent cross reactivity (% CR) of Dextran T-40 with other Dextran molecular weights is shown in the table below.

Dextran Molecular Weights	Sigma Cat#	% Cross-Reactivity
T-40	31389	100
T-6	31388	25
T-100	01984	121
T-450	31392	107

### Wash Solution Preparation

1. Measure 1 L of laboratory quality distilled or deionized water and transfer to a clean container with a tight-fitting lid.
2. Transfer the contents of 1 Wash Concentrate packet to the container, swirl to mix.
3. Transfer the Wash Solution to a wash bottle to use in the assay.

### Fresh Beet Juice Sample Preparation

1. Centrifuge juice samples for 5 minutes at 12,000 X g to clarify.
2. Prepare a 1:100 dilution by diluting 100 µL of the clarified juice sample with 9.9 mL of laboratory quality distilled or deionized water and mix thoroughly.

### Assay Procedure

1. Allow kit components and the sample extract(s) to reach room temperature prior to running the test.
2. Place the appropriate number of test tubes into a tube rack. Label the tubes one inch from the top with the control concentration/identification or sample identification. Be sure to re-seal unused tubes in the zip-lock bag with the desiccant to limit exposure to moisture.
3. Dispense **250 µL of Dextran HRP** into each tube.
4. Dispense **250 µL of Controls and Samples into the appropriate tubes**. Be sure to use a clean pipette tip for each solution to avoid cross contamination.
5. Dispense **250 µL of Antibody** into each tube.
6. Gently shake the tubes for 30 seconds using a back-and-forth motion and incubate for **20 minutes** at room temperature.
7. Decant the contents of the tubes into an appropriate waste container. Fill the tubes to overflowing with wash solution and then decant. Repeat this wash step three times for a total of four washes. Following the last wash, tap the inverted tubes onto absorbent paper to remove excess wash solution.
8. Dispense **500 µL of Substrate** into each tube.
9. Gently shake the tubes for 30 seconds using a back-and-forth motion and incubate for **20 minutes** at room temperature.
10. Dispense **500 µL of Stop Solution** into each tube in the same order as the Substrate.
11. Gently shake the tubes for 30 seconds using a back-and-forth motion.
12. Carefully wipe the optical surface with a soft, lint-free wipe. Measure and record the absorbance (Optical Density; OD) of each tube at 450 nm using a tube reader within 10 minutes of stopping the assay. Be sure to blank the reader with DI water prior to measuring.
13. Dispose of used test tubes in an appropriate waste container.

### Quality Control (QC) Criteria

- The average absorbance of the negative control replicates must be  $\geq 0.5$ .
- The average absorbance of control replicates must have a coefficient of variation (%CV) < 15%.
- The average absorbance of sample replicates must have a coefficient of variation (%CV) < 20%.