Flatulence: Experimental Description & Data

Carbohydrates, Digestion & Enzymes

Background:

Stock solutions were prepared by soaking ~50g of raw RFO containing produce, which were selected from the Table: *Foods Associated with Flatulence*, with 100mL of deionized water at 25 ^{o}C for 12h. Each of the stock solutions served as the initial stock substrate $[S]_o$ solution that was diluted and incubated with active enzymes: α-galactosidase and sucrase, which are contained in the commercial product liquid *Beano*. The catalyzed reactions produce glucose and fructose by converting the Raffinose Family of Oligosaccharides (RFOs) to the simple sugars, glucose and fructose in two steps.

RFO Oligosaccharides + H_2O $\xrightarrow{\alpha\text{-galactosidase}}$ galactose + sucrose

Sucrose + H_2O $\xrightarrow{\text{sucrase}}$ glucose + fructose

Procedure: *(The following procedure was generally followed with adaptation for fresh produce, eg. broccoli.)*

5mL of each of the respective solutions was placed in a vial and labeled $[S]_o$. A second 5mL aliquot was diluted with deionized water to 10mL and labeled $0.5[S]_o$. 5mL of this solution was diluted to 10mL and placed in a third vial labeled as $0.25[S]_o$. The vials were sealed and placed in a constant temperature water bath for 20-30 minutes at either 25 ^{o}C or 35 ^{o}C. 10μL of liquid *Beano* was added to each vial and the time recorded. One drop from each substrate vial was tested every 5-10 minutes over a period of 40-50 minutes using a glucometer. (Glucometers are used by diabetics to monitor their blood sugar levels.) The cost per data point (analysis) is ~$0.50.

Near the end of the test period, 4-5 drops of 0.1 M $HCl_{(aq)}$ were added to an aliquot of $[S]_o$. Placed in a constant temperature bath for ~5min, then tested.

Data:

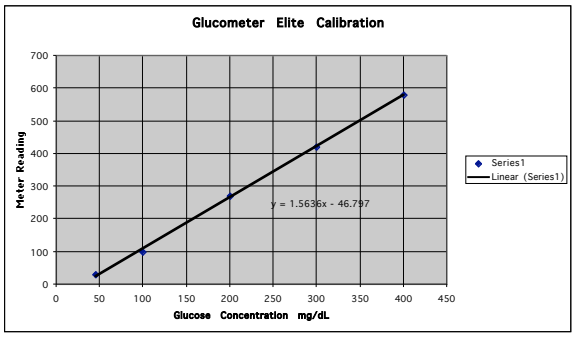
Sugar Source	*Concentration*	*Reading*	*Temperature*
	[S]o	Glucometer	
	[S]o =100mL deionized H_2O Extracts of 50g of produce		
	time (min)	(mg/dL)	(oC)
Split Green Peas	[S]o		25
	5	110	
	15	402	
	25	526	
	35	582	
	45	574	
Added to a separate aliquot	+ 0.1M HCl		
@ 25 min; incubated & tested	25	110	
	0.25[S]o		25
	10	68	
	20	131	
	30	151	
	40	149	
	0.5[S]o		35
	3	83	
	18	345	
	28	370	
	41	363	
	63	360	
Red Kidney Beans	0.5[S]o		25
	3	32	
	14	41	
	17	53	
	40	52	
	0.5[S]o		35
	12	63	
	28	74	
	36	69	
	45	74	

PART I : (***Clearly*** answer the following questions on the form provided and turn in. Can be done in partnerships of no larger than three members per group.)

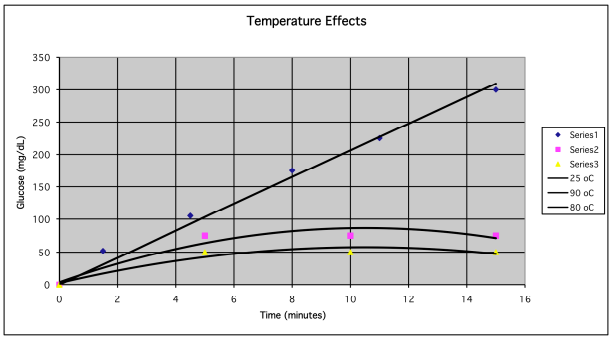
1. Draw a Haworth structure for sucrose.
2. Draw Fisher formulas for the un-cyclized forms and Haworth structures for the cylclized forms of D-glucose and D-fructose. Label the cyclized forms as either the α-anomer or β-anomer.
3. What are the respective molecular formulas and Molar Masses of a) verbascose, b) stachyose, and c) raffinose?
4. How many grams of glucose and fructose would be produced by complete hydrolysis of 5 mmol of a) verbascose, b) stachyose, and c) raffinose respectively?
5. How much energy (kJ) is produced from the complete metabolic oxidation of 5 mmol of glucose to CO_2 and H_2O?

PART II : (***Clearly*** answer the following questions on the form provided and turn in. Can be done in partnerships of no larger than three members per group.)

1. Glucometers are calibrated for blood chemistry and not for simple aqueous solutions. Therefore, using the calibration graph that follows, convert the *Glucometer Elite*™ data readings for split green peas to respective concentrations of glucose. Normalize the glucose data/concentrations of 0.25[So] and 0.50[So] to the [So]'s concentration/glucose data. Clearly graph glucose versus time for each of the trials on the same graph. Label each.



2. The *Glucometer Elite*™ measures the concentration of only the β-anomer of D-glucose. A computer (logic) chip processes the input to determine the overall concentration of glucose. The calibration curve that was produced used glucose solutions that were prepared 12h before analysis. Draw Haworth structures for the α-anomer and β-anomer of D-glucose.
3. Explain how mutarotation of the α-anomer and β-anomer of D-glucose and K_{eq} need to be considered in programming and building the computer (logic) chip in the *Glucometer Elite*™.
4. Do you think that *Beano* is just as effective *in vivo* as in these tests? Explain within the context of stomach acidity and oral administration.
5. What volume of gas could gastrointestinal bacteria theoretically produce from complete fermentation of 100g of red beans that were eaten by Dr. R. in his favorite Los Panchos' burrito? Clearly state your assumptions and show your calculation. (A review of General Chem gas laws would be a good place to begin. You can assume that Los Pancho's would produce nothing less than "ideal" gases.)
6. Certain animals obtain food energy and fiber from the digestion of cellulose (tree bark). We cannot. If *Beano* were added to tree bark, could this combination be included as a healthy menu item in DVC's cafeteria? Briefly explain your answer.
7. Refer to the graph below. Would *Beano* work if added to foods before cooking? Estimate the optimum temperature for *Beano* performance. Explain your answers.



Raw Lab Data:

Glucometers are calibrated for blood chemistry and not for simple aqueous solutions. Therefore, using the calibration graph that follows, convert the *Glucometer Elite*™ data readings for split green peas to respective concentrations of glucose. Place them in a separate table and graph the concentrations versus time for each of the 3 substrate trials on the same graph. Answer the Post-lab questions.

Sugar Source	*Concentration*	*Reading*	*Temperature*
	[S]o	Glucometer	
	[S]o =100mL deionized H_2O Extracts of 50g of produce		
	time (min)	(mg/dL)	(oC)
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