

Growth curve data for Modelling

TUESDAY, 10/1/2019

Aim: To determine the growth rate of *Vibrio natriegens* in varying salt concentrations.

The growth curve data of *Vibrio natriegens* in different media was determined by using;

1. LB, BHI and MM media with varying NaCl concentrations.
2. LB and BHI with V2 and V3 salts.

Components used:

- i. 2x LB media
- ii. 2x BHI media
- iii. 2x Mineral media
- iv. 5M NaCl
- v. 10x V2 salts (204mM NaCl, 4.2mM KCl, 23.14mM MgCl₂)
- vi. 10x V3 salts (475mM NaCl, 9.7mM KCl, 54mM MgCl₂)
- vii. *Vibrio natriegens* grown in LBV overnight at 37 degrees, 200rpm.

Layout of the Plate reader used:

Well1												
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.5g/L (NaCl)	0.5g/L (NaCl)	0.5g/L (NaCl)	5g/L (NaCl)	5g/L (NaCl)	5g/L (NaCl)	10g/L (NaCl)	10g/L (NaCl)	10g/L (NaCl)	15g/L (NaCl)	15g/L (NaCl)	15g/L (NaCl)
B	20g/L (NaCl)	20g/L (NaCl)	20g/L (NaCl)	22.5g/L (NaCl)	22.5g/L (NaCl)	22.5g/L (NaCl)	25g/L (NaCl)	25g/L (NaCl)	25g/L (NaCl)	27.5g/L (NaCl)	27.5g/L (NaCl)	27.5g/L (NaCl)
C	30g/L (NaCl)	30g/L (NaCl)	30g/L (NaCl)	32.5g/L (NaCl)	32.5g/L (NaCl)	32.5g/L (NaCl)	35g/L (NaCl)	35g/L (NaCl)	35g/L (NaCl)	37.5g/L (NaCl)	37.5g/L (NaCl)	37.5g/L (NaCl)
D	40g/L (NaCl)	40g/L (NaCl)	40g/L (NaCl)	45g/L (NaCl)	45g/L (NaCl)	45g/L (NaCl)	LBV2	LBV2	LBV2	LBV3	LBV3	LBV3
E	0.5g/L (NaCl)	0.5g/L (NaCl)	0.5g/L (NaCl)	5g/L (NaCl)	5g/L (NaCl)	5g/L (NaCl)	10g/L (NaCl)	10g/L (NaCl)	10g/L (NaCl)	15g/L (NaCl)	15g/L (NaCl)	15g/L (NaCl)
F	20g/L (NaCl)	20g/L (NaCl)	20g/L (NaCl)	22.5g/L (NaCl)	22.5g/L (NaCl)	22.5g/L (NaCl)	25g/L (NaCl)	25g/L (NaCl)	25g/L (NaCl)	27.5g/L (NaCl)	27.5g/L (NaCl)	27.5g/L (NaCl)
G	30g/L (NaCl)	30g/L (NaCl)	30g/L (NaCl)	32.5g/L (NaCl)	32.5g/L (NaCl)	32.5g/L (NaCl)	35g/L (NaCl)	35g/L (NaCl)	35g/L (NaCl)	37.5g/L (NaCl)	37.5g/L (NaCl)	37.5g/L (NaCl)
H	40g/L (NaCl)	40g/L (NaCl)	40g/L (NaCl)	45g/L (NaCl)	45g/L (NaCl)	45g/L (NaCl)	BHIV2	BHIV2	BHIV2	BHIV3	BHIV3	BHIV3

Row A-D: LB media

Components per well:

1. 100uL 2x LB
2. 10uL culture
3. varying concentrations of NaCl solution=x
4. MilliQ= 90-x

Total= 200uL

For LBV2 and LBV3 wells:

1. 100uL 2x LB
2. 10uL culture
3. 20uL 10x V2 and 20uL V3 separately
4. MilliQ= 90-x

Total= 200uL

	A	B
1	NaCl conc. (g/L)	Amount of 5M NaCl to be added to media (uL)= x
2	0.5	0
3	5	3.42
4	10	6.84
5	15	10.26
6	20	13.68
7	22.5	15.39
8	25	17.1
9	27.5	18.81
10	30	20.52
11	32.5	22.23
12	35	23.94
13	37.5	25.65
14	40	27.36
15	45	30.78

Row E-F: BHI media

1. 100uL 2x BHI
2. 10uL culture
3. varying concentrations of NaCl solution=x
4. MilliQ= 90-x

Total= 200uL

For BHIV2 and BHIV3 wells:

1. 100uL 2x BHI
2. 10uL culture
3. 20uL 10x V2 and 20uL V3 separately
4. MilliQ= 90-x

Total= 200uL

The 96 well plate with samples was inserted in the plate reader and readings were obtained at every 15 minute interval for 8 hours.

 Growth curve vibrio (LB BHI diff NaCl conc).xlsx**THURSDAY, 10/17/2019**

Aim: To determine the optimal concentration of glucose required to obtain optimal growth of *Vibrio natriegens* in minimal media.

Materials required: Sodium glucuronate 2g/L, MgSO₄ · 7H₂O 1-3 g/L';