



Pendahuluan

- Perhitungan diperlukan untuk mengetahui jumlah bakteri atau mikroorganisme yang akan digunakan dalam proses produksi sehingga memperkecil kegagalan proses.
  - Perhitungan dapat dilakukan dengan beberapa cara

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Generation Number	Number of Cells	$\log_{10}$ of Number of Cells
0	1	0
$5(2^5) =$	32	1.51
$10(2^{10}) =$	1,024	3.01
$15(2^{15}) =$	32,768	4.52
$16(2^{16}) =$	65,536	4.82
$17(2^{17}) =$	131,072	5.12
$18(2^{18}) =$	262,144	5.42
$19(2^{19}) =$	524,288	5.72
$20(2^{20}) =$	1,048,576	6.02

Figure 6.12b

Jika 100 sel ditumbuhkan selama 5 jam dan menghasilkan 1.720.320 sel:

$$\text{Number of generations} = \frac{\text{Log number of cells (end)} - \text{Log number of cells (beginning)}}{0.301}$$

$$\text{Jumlah generasi} = (\log 1.720.320 - \log 100)/0,301 = 14$$

$$\text{Generation time} = \frac{60 \text{ min} \times \text{hours}}{\text{Number of generations}} = 21 \text{ minutes/generasi}$$

$$\text{Waktu generasi} = (60 \times 5)/14 = 21 \text{ menit/generasi}$$

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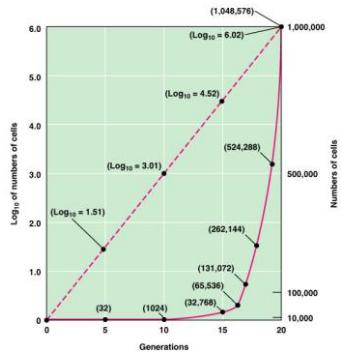
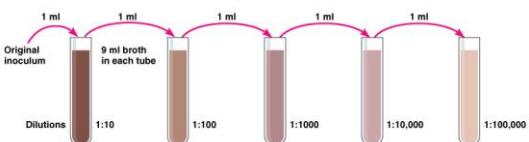


Figure 6.13

## Pengukuran langsung pertumbuhan mikroba

- Plate Counts: Mendasarkan pada seri pengenceran sampel

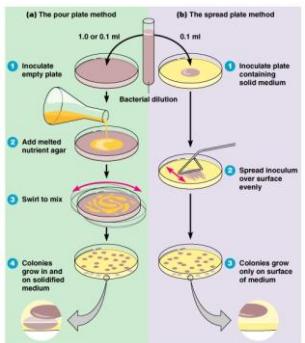


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Figure 6.15, top portion

### Plate Count

- Inokulasi cawan Petri dari seri pengenceran

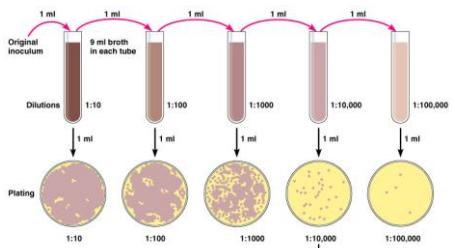


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Figure 6.16

## Plate Count

- Setelah inkubasi, hitung koloni pada cawan yang memiliki jumlah 25-250 koloni (CFU)

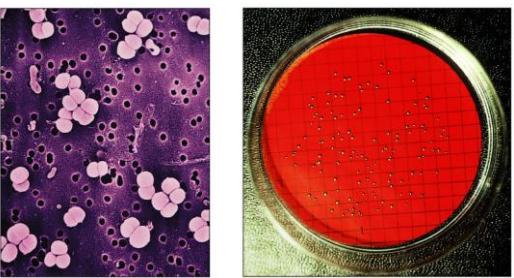


**Calculation:** Number of colonies on plate  $\times$  reciprocal of dilution of sample = number of bacteria/ml  
 (For example, if 32 colonies are on a plate of  $1/10,000$  dilution, then the count is  $32 \times 10,000 = 320,000/\text{ml}$  in sample.)

Figure 6.15

## Pengukuran langsung pertumbuhan mikrobia

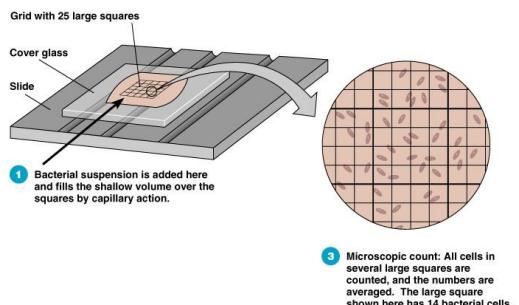
- **Filtrasi**



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Figure 6.17a, b

## Pengukuran langsung pertumbuhan mikrobia



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Figure 6.19

## Pengukuran langsung pertumbuhan mikrobia

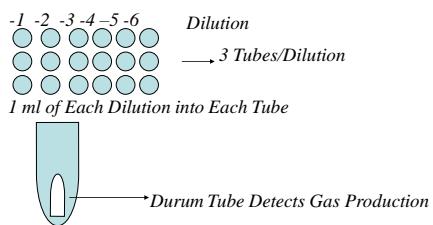
- Hitung langsung dengan Mikroskop

$$\text{Number of bacteria/ml} = \frac{\text{number of cells counted}}{\text{volume of area counted}}$$

$$\frac{14}{8 \times 10^{-7}} = 17,500,000$$

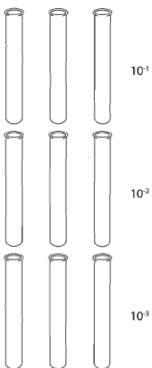
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## Most Probable Number (MPN)

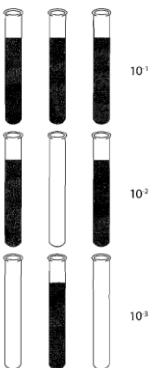


POSITIVE TUBES Have GROWTH and GAS PRODUCTION

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**Figure 1:** Three-tube design for MPN (unincubated).

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**Figure 2:** Three-tube design for MPN (incubated).


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**MPN - Continued**

- Recording Results  
-1 -2 -3 -4 -5 -6  
3 3 2 1 0 0 - # of (+) Tubes
- Objective is to “DILUTE OUT” the organism
- Select Most Dilute Sample with All Positive and Take Next Two
  - 3,2,1 – Our Combination
- Determine MPN Using MPN Table

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**MPN**

- Calculations
  - If 3, 2, 1 Then Result is: 150/ml or g
  - This is IF we used the the -1, -2, -3 Dilution
  - WE used the -2,-3,-4 Dilutions so we must multiply by 10
- OUR RESULT
  - 1500 cfu/g or ml
  - $1.5 \times 10^3$  cfu/g or ml

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## MPN

- If none of the tubes have 3 positive, take most dilute with a positive and take next two ABOVE it
- Examples
  - 1 -2 -3 -4 -5 -6
  - a. 3 3 3 **3 1 1**
  - b. **2 2 1** 0 0 0
  - c. 3 3 2 1 1 0 ??????

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## MPN

- For C, use 3 2 2
  - There is a tube that has a positive result “below” the -4 tube (most dilute tube used in the result)
- If a tube that is more dilute than the tubes in the 3 tube combination has a positive result then combine that number with the last tube of the 3 tube combination!!

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Example	Sample quantities (g or ml) <sup>1</sup>					Reported positive values	MPN estimate/ g or ml
	10	1	0.1	0.01	0.001		
a	3/3 <sup>2</sup>	<b>3/3</b>	<b>2/3</b>	<b>0/3</b>	0/3	3-2-0	9.3
b	3/3	3/3	<b>3/3</b>	<b>2/3</b>	<b>0/3</b>	3-2-0	93.
c	0/3	<b>0/3</b>	<b>1/3</b>	<b>0/3</b>	0/3	0-1-0	0.31
d	3/3	<b>3/3</b>	<b>2/3</b>	<b>1/3</b>	1/3	3-2-2	21.
e	3/3	3/3	<b>3/3</b>	<b>3/3</b>	<b>3/3</b>	3-3-3	>1100

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## Pengukuran langsung pertumbuhan mikroba

- Tabung ganda (uji MPN)
- Hitung tabung positif dan bandingkan dengan tabel MPN.

Combination of Positives	MPN Index/ 100 ml	95% Confidence Limits	
		Lower	Upper
4-2-0	22	9	56
4-2-1	26	12	65
4-3-0	27	12	67
4-3-1	33	15	77
4-4-0	34	16	80
5-0-0	23	9	86
5-0-1	30	10	110
5-0-2	40	20	140
5-1-0	30	10	120
5-1-1	50	20	150
5-1-2	60	30	180
5-2-0	50	20	170
5-2-1	70	30	210
5-2-2	90	40	250
5-3-0	80	30	250
5-3-1	110	40	300
5-3-2	140	60	360

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Figure 6.18b

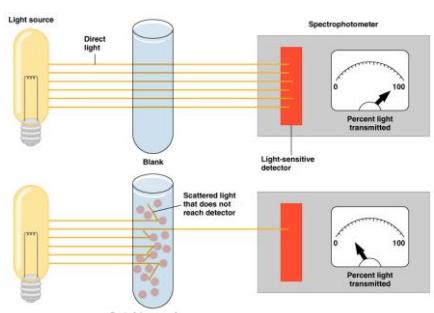
## Dengan luas bidang pandang

- Ratakan suspensi bakteri 0,1 mL pada gelas objek seluas (1X4) cm persegi
- Amati jumlah bakteri per luas bidang pandang
- Ukur luas bidang pandang
- Tentukan jumlahnya

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## Pendugaan Jumlah Bakteri dengan Metode Tidak Langsung

- Turbiditas



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Figure 6.20

### Pendugaan Jumlah Bakteri dengan Metode Tidak Langsung

- Aktivitas metabolismik
- Berat kering

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