Fisica do Corpo Humano (4300325)

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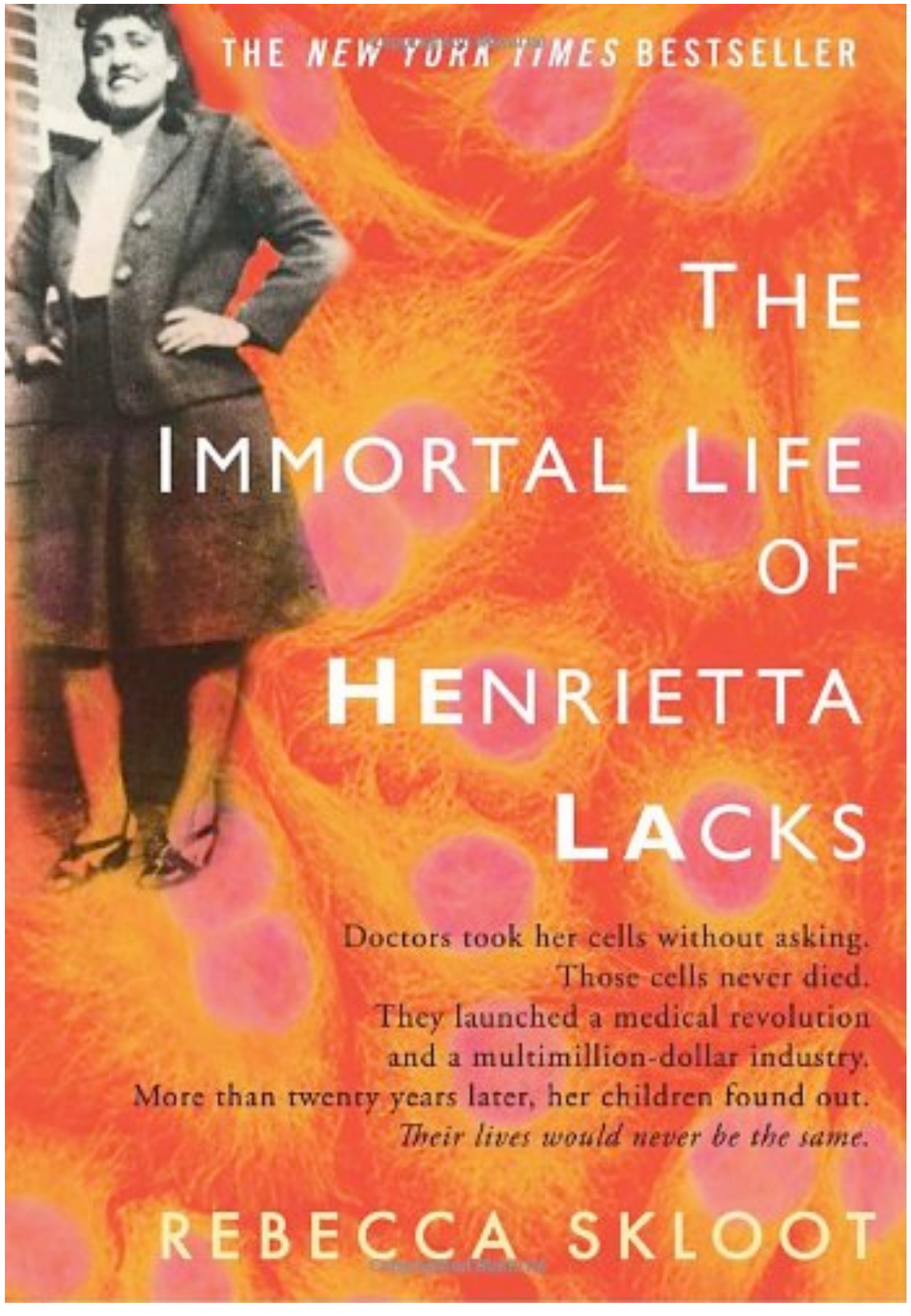
C01

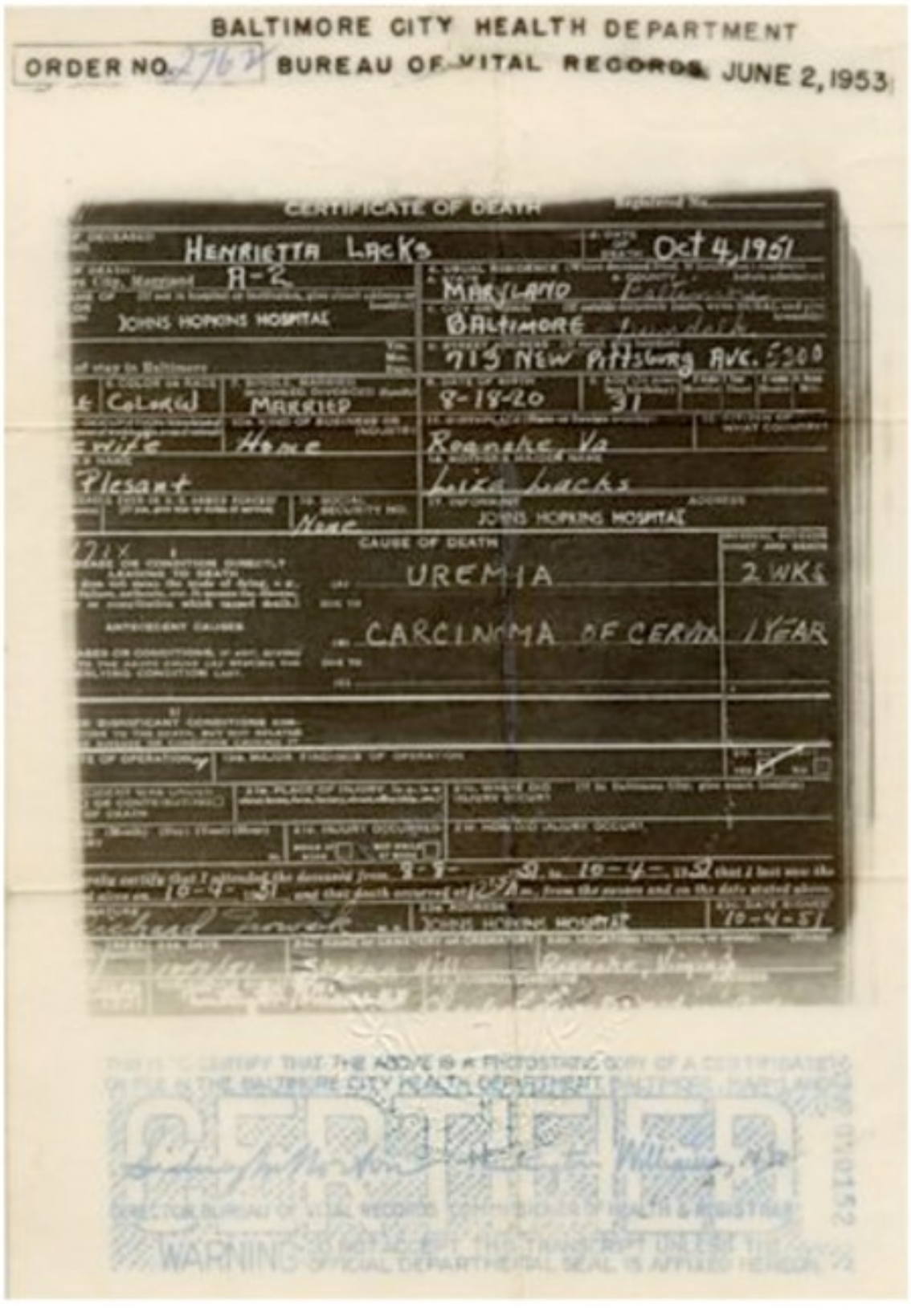
Elementos Celulares e Microscopia
Aula 14

Teoria Celular

Em 1838, dois cientistas cujas pesquisas centradas na inspeção microscópica dos seres vivos propôs a **Teoria Celular**. **Matthais Schleiden** trabalhou com plantas e pode ter vindo com a maior parte da hipótese de que **Theodor Schwann** estendeu aos animais. O Teoria celular, como é aplicado hoje, diz:

1. Tudo o que vivo é composta de pelo menos uma célula.
2. As células só pode ser feita a partir de células existentes, relacionados.
3. O menor única coisa que pode ser considerado vivo é uma célula.
4. As células de todos os seres vivos são mais parecidos do que diferentes.


THE NEW YORK TIMES BESTSELLER
THE IMMORTAL LIFE OF HENRIETTA LACKS
Doctors took her cells without asking.
Those cells never died.
They launched a medical revolution
and a multimillion-dollar industry.
More than twenty years later, her children found out.
Their lives would never be the same.
REBECCA SKLOOT


BALTIMORE CITY HEALTH DEPARTMENT
ORDER NO. 2767 BUREAU OF VITAL RECORDS JUNE 2, 1953
CERTIFICATE OF DEATH
HENRIETTA LACKS
Oct 4, 1951
COLORED
MARRIED
8-18-20
31
Clover
Pleasant
JOHNS HOPKINS HOSPITAL
CAUSE OF DEATH
UREMIA
2 WKS
CARCINOMA OF CERVIX
1 YEAR
COPIED

Henrietta Lacks morreu de câncer cervical em 1951. Antes de morreu um cirurgião retirou amostras desse tumor e colocou em um prato de petri. Cientistas buscavam manter cultura de células humanas a décadas, mas eventualmente essas células morriam. As células do tumor de Henrietta eram diferente: elas duplicavam a cada 24 horas e nunca paravam de crescer. Elas se tornaram as primeiras células humanas imortais crescidas em laboratório.

1/8/1920 - 4/10/1951

Timeline | **The development of human cancer cell lines**

Ross G. Harrison develops the 'hanging drop culture' to study frog nerve-cell growth.

Wilton R. Earle and George Gey generate a rodent continuous cell line.

Klaus H. Rothfels and colleagues show interspecies cross-contamination.

Walter Nelson-Rees shows widespread HeLa cross-contamination.

Cross-contaminated cell lines are used at record levels.

1907 1910 1940 1951 1958 1967 1974 1990 2002

Montrose T. Burrows and Alexis Carrel grow chick embryo cells in tissue culture.

George and Margaret Gey and Mary Kubicek develop HeLa, the first human cancer continuous cell line.

Stan Gartler shows intraspecies cross-contamination.

Dennis Gilbert, Stephen O'Brien and colleagues apply multilocus DNA fingerprinting to cell-line authentication.

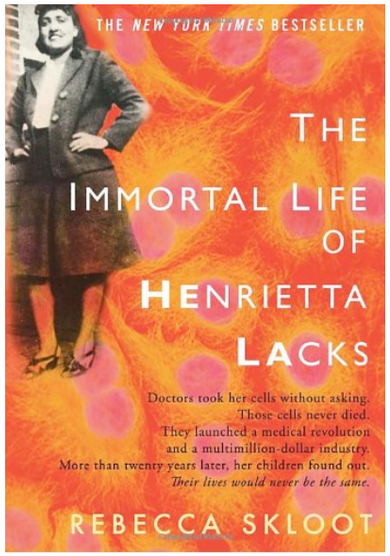
George Gay, diretor do laboratório no Johns Hopkins Hospital, onde ocorreu a primeira cultura de células bem sucedida. Células HeLa.

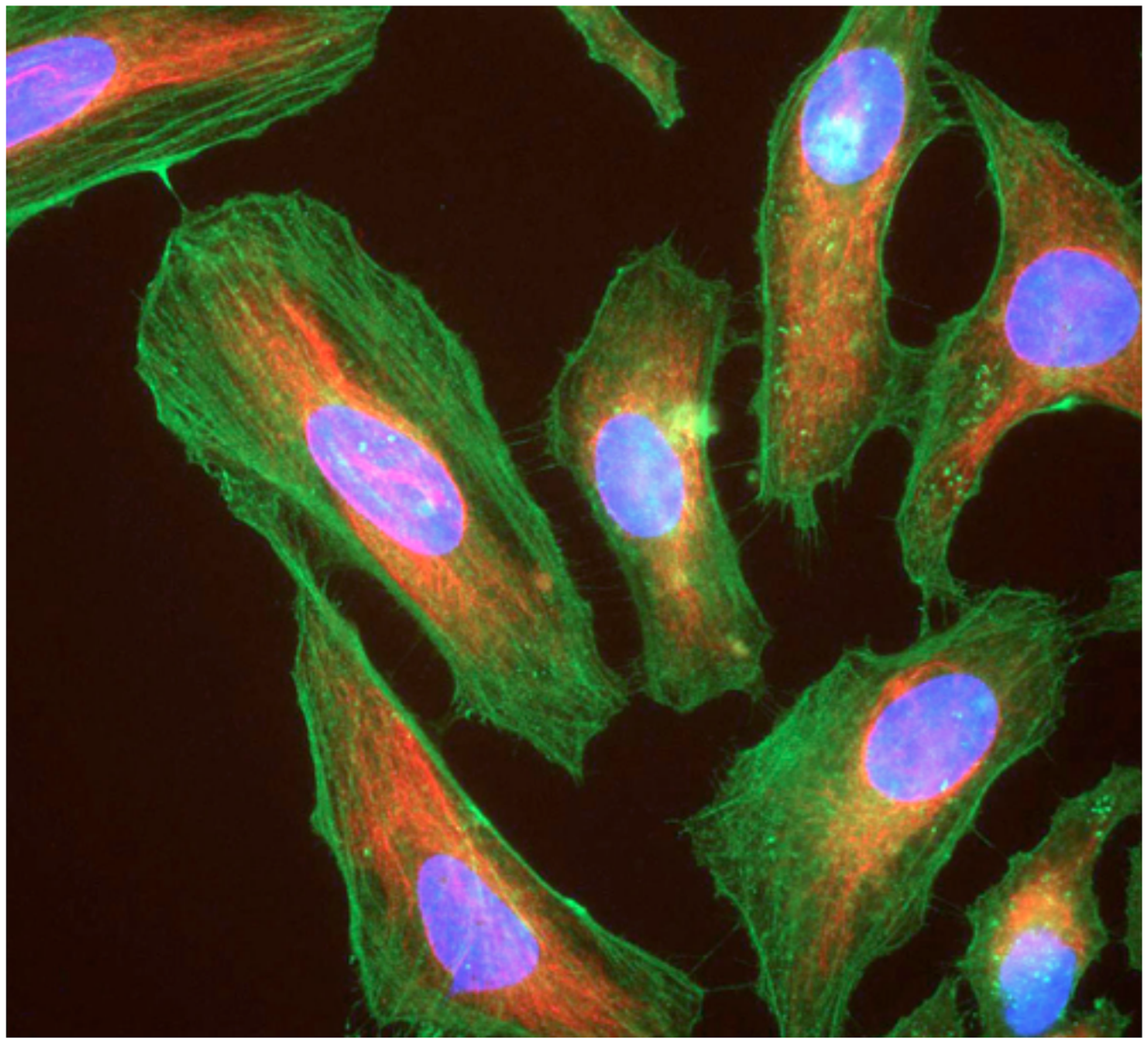
The development of human cancer cell lines, John R. Masters, Nature Reviews Cancer 2, 315-319 (April 2002) doi:10.1038/nrc775

As **células HeLa** foram utilizadas por **Jonas Salk** para **testar** a primeira **vacina** contra a **poliomielite** em 1953. As células HeLa eram facilmente infectadas por poliomielite, fazendo com que as células infectadas morresem [1]. Um grande volume de células HeLa foi necessário para os testes da vacina contra a pólio Salk, o que levou a Fundação Nacional para a Paralisia Infantil (NFIP) encontrar um local capaz de fazer uma **produção em massa de células HeLa** [2]. Na primavera de 1953, a fabrica de cultura de células foi fundada na Universidade de Tuskegee para abastecer Salk, bem como de outros laboratórios, com células HeLa [3]. Menos de um ano depois, a **vacina Salk estava pronta** para testes em humanos.

[1] Scherer, W. F.; Syverton, JT; Gey, GO (1953). "*Studies On The Propagation In Vitro Of Poliomyelitis Viruses: Iv. Viral Multiplication In A Stable Strain Of Human Malignant Epithelial Cells (Strain Hela) Derived From An Epidermoid Carcinoma Of The Cervix*". Journal of Experimental Medicine 97 (5): 695–710.
[2] The development of human cancer cell lines, John R. Masters, Nature Reviews Cancer 2, 315-319 (April 2002) doi:10.1038/nrc775
[3] Turner, Timothy (2012). "Development of the Polio Vaccine: A Historical Perspective of Tuskegee University's Role in Mass Production and Distribution of HeLa Cells". Journal of Health Care for the Poor and Underserved 23 (4a): 5–10. doi:10.1353/hpu.2012.0151

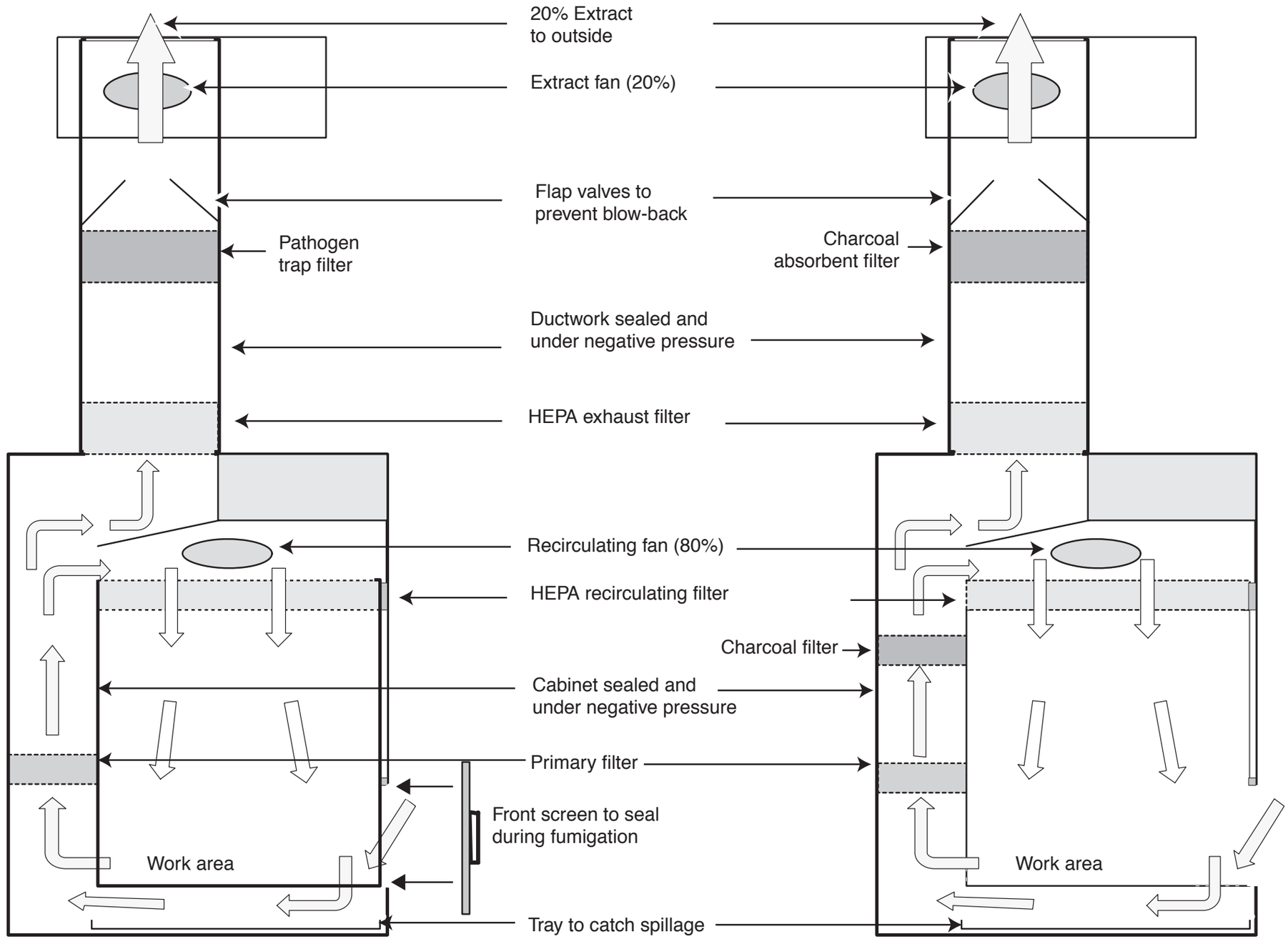
“Se você pudesse acumular todas as células HeLa já cultivadas em uma escala, elas pesariam mais de 50 milhões de toneladas métricas, o equivalente a pelo menos 100 Empire State Buildings. Outro cientista calculou que, se você colocasse todas as células HeLa já cultivadas lado a lado, elas embrulhariam a Terra pelo menos três vezes, abrangendo mais de 350 milhões pés.”





HeLa cells stained with antibody to actin (green), vimentin (red) and DNA (blue). Image courtesy of EnCor Biotechnology Inc.

Gabinete de Segurança



(a) CLASS II SAFETY CABINET

(b) CHEMICAL SAFETY CABINET

Frasco de cultura

Fig. 8.3. Petri Dishes. Illustrated are dishes of 3.5-cm, 5-cm, and 9-cm diameter. Square Petri dishes are also available, with dimensions 9 × 9 cm. A grid pattern can be provided to help in scanning the dish—for example, in counting colonies—but can interfere with automatic colony counting.

Fig. 8.4. Plastic Flasks. Sizes illustrated are 10 and 25 cm^2 (Falcon, B-D Biosciences), 75 cm^2 (Corning), and 185 cm^2 (Nalge Nunc) (*see* Table 8.1 for representative sizes and capacities).

Fig. 8.8. Venting Flasks. Gas-permeable cap on 10-cm^2 flask (Falcon, B-D Biosciences).

Meio de cultura

TABLE 9.3. Frequently Used Media

Component	MEM	DMEM	F12	DMEM/ F12	αMEM	CMRL 1066	RPMI 1640	M199	L15	McCoy's 5A	Fischer	MB 752/1
Amino acids												
L-alanine			1.0E-04	5.0E-05	2.8E-04	2.8E-04		2.8E-04	2.5E-03	1.5E-04		
L-arginine	6.0E-04	4.0E-04	1.0E-03	7.0E-04	6.0E-04	3.3E-04	1.1E-03	3.3E-04	2.9E-03	2.0E-04	7.1E-05	3.6E-04
L-asparagine			1.0E-04	5.0E-05	3.3E-04	3.8E-04	1.7E-03	3.0E-04	7.6E-05			
L-aspartic acid			1.0E-04	5.0E-05	2.3E-04	2.3E-04	1.5E-04	2.3E-04		1.5E-04		4.5E-04
L-cysteine			2.0E-04	1.0E-04	5.7E-04	1.5E-03		5.6E-07	9.9E-04	2.0E-04		5.0E-04
L-cystine	1.0E-04	2.0E-04		1.0E-04	1.0E-04	8.3E-05	2.1E-04	9.9E-05			9.9E-05	6.3E-05
L-glutamic acid			1.0E-04	5.0E-05	5.1E-04	5.1E-04	1.4E-04	4.5E-04		1.5E-04		1.0E-03
L-glutamine	2.0E-03	4.0E-03	1.0E-03	2.5E-03	2.0E-03	6.8E-04	2.1E-03	6.8E-04	2.1E-03	1.5E-03	1.4E-03	2.4E-03
Glycine		4.0E-04	1.0E-04	2.5E-04	6.7E-04	6.7E-04	1.3E-04	6.7E-04	2.7E-03	1.0E-04		6.7E-04
L-histidine	2.0E-04	2.0E-04	1.0E-04	1.5E-04	2.0E-04	9.5E-05	9.7E-05	1.0E-04	1.6E-03	1.0E-04	3.9E-04	8.3E-04
L-hydroxy-proline						7.6E-05	1.5E-04	7.6E-05		1.5E-04		
L-isoleucine	4.0E-04	8.0E-04	3.0E-05	4.2E-04	4.0E-04	1.5E-04	3.8E-04	1.5E-04	9.5E-04	3.0E-04	5.7E-04	1.9E-04
L-leucine	4.0E-04	8.0E-04	1.0E-04	4.5E-04	4.0E-04	4.6E-04	3.8E-04	4.6E-04	9.5E-04	3.0E-04	2.3E-04	3.8E-04
L-lysine HCl	4.0E-04	8.0E-04	2.0E-04	5.0E-04	4.0E-04	3.8E-04	2.2E-04	3.8E-04	5.1E-04	2.0E-04	2.7E-04	1.3E-03
L-methionine	1.0E-04	2.0E-04	3.0E-05	1.2E-04	1.0E-04	1.0E-04	1.0E-04	1.0E-04	5.0E-04	1.0E-04	6.7E-04	3.4E-04
L-phenylalanine	2.0E-04	4.0E-04	3.0E-05	2.2E-04	1.9E-04	1.5E-04	9.1E-05	1.5E-04	7.6E-04	1.0E-04	4.1E-04	3.0E-04
L-proline			3.0E-04	1.5E-04	3.5E-04	3.5E-04	1.7E-04	3.5E-04		1.5E-04		4.3E-04
L-serine		4.0E-04	1.0E-04	2.5E-04	2.4E-04	2.4E-04	2.9E-04	2.4E-04	1.9E-03	2.5E-04	1.4E-04	
L-threonine	4.0E-04	8.0E-04	1.0E-04	4.5E-04	4.0E-04	2.5E-04	1.7E-04	2.5E-04	2.5E-03	1.5E-04	3.4E-04	6.3E-04
L-tryptophan	4.9E-05	7.8E-05	1.0E-05	4.4E-05	4.9E-05	4.9E-05	2.5E-05	4.9E-05	9.8E-05	1.5E-05	4.9E-05	2.0E-04
L-tyrosine	2.0E-04	4.0E-04	3.0E-05	2.1E-04	2.3E-04	2.2E-04	1.1E-04	2.2E-04	1.7E-03	1.2E-04	3.3E-04	2.2E-04
L-valine	4.0E-04	8.0E-04	1.0E-04	4.5E-04	3.9E-04	2.1E-04	1.7E-04	2.1E-04	8.5E-04	1.5E-04	6.0E-04	5.6E-04
Vitamins												
p-Aminobenzoic acid						3.6E-07	7.3E-06	3.6E-07		7.3E-06		
L-Ascorbic acid					2.5E-04	2.8E-04		2.8E-07		3.2E-06		9.9E-05
Biotin			3.0E-08	1.5E-08	4.1E-07	4.1E-08	8.2E-07	4.1E-08		8.2E-07	4.1E-08	8.2E-08
Calciferol								2.5E-07				
Choline chloride	7.1E-06	2.9E-05	1.0E-04	6.4E-05	7.1E-06	3.6E-06	2.1E-05	3.6E-06	7.1E-06	3.6E-05	1.1E-05	1.8E-03
Folic acid	2.3E-06	9.1E-06	2.9E-06	6.0E-06	2.3E-06	2.3E-08	2.3E-06	2.3E-08	2.3E-06	2.3E-05	2.3E-05	9.1E-07
myo-Inositol	1.1E-05	4.0E-05	1.0E-04	7.0E-05	1.1E-05	2.8E-07	1.9E-04	2.8E-07	1.1E-05	2.0E-04	8.3E-06	5.6E-06
Menadione								6.9E-08				
Nicotinamide	8.2E-06	3.3E-05	3.3E-07	1.7E-05	8.2E-06	2.0E-07	8.2E-06	2.0E-07	8.2E-06	4.1E-06	4.1E-06	8.2E-06
Nicotinic acid								2.0E-07		4.1E-06		
D-Ca pantothenate	4.2E-06	1.7E-05	2.0E-06	9.4E-06	4.2E-06	4.2E-08	1.1E-06	4.2E-08	4.2E-06	8.4E-07	2.1E-06	4.2E-06
Pyridoxal HCl	4.9E-06	2.0E-05	3.0E-07	1.0E-05	4.9E-06	1.2E-07		1.2E-07		2.5E-06	2.5E-06	
Pyridoxine HCl			3.0E-07	1.5E-07		1.2E-07	4.9E-06	1.2E-07		2.4E-06		4.9E-06
Riboflavin	2.7E-07	1.1E-06	1.0E-07	5.8E-07	2.7E-07	2.7E-08	5.3E-07	2.7E-08	1.9E-07	5.3E-07	1.3E-06	2.7E-06
Thiamin	3.0E-06	1.2E-05	1.0E-06	6.4E-06	3.0E-06	3.0E-08	3.0E-06	3.0E-08	2.4E-06	5.9E-07	3.0E-06	3.0E-05
Thiamin mono PO_4										4.8E-06		
α-Tocopherol								2.3E-08				
Retinol acetate								3.5E-07				
Vitamin B_{12}			1.0E-06	5.0E-07	1.0E-06		3.7E-09					1.5E-07
Antioxidants												
Glutathione						3.0E-05	3.0E-06	1.5E-07		1.5E-06		4.5E-05
Inorganic salts												
$CaCl_2$	1.8E-03	1.8E-03	3.0E-04	1.1E-03	1.8E-03	1.8E-03		1.3E-03	1.3E-03	9.0E-04	6.2E-04	8.2E-04
KCl	5.3E-03	5.3E-03	3.0E-03	4.2E-03	5.3E-04	5.3E-03	5.3E-03	5.3E-03	5.3E-03	5.3E-03	5.3E-03	2.0E-03
KH_2PO_4								4.4E-04	4.4E-04			5.9E-04
$MgCl_2$					1.2E-01							1.2E-03
$MgSO_4$	8.1E-04	8.1E-04		4.0E-04	8.1E-04	8.1E-04	4.0E-04	8.1E-04	1.6E-03	8.1E-04	4.9E-04	8.1E-04
NaCl	1.2E-01	1.1E-01	1.3E-01	1.2E-01		1.2E-01	1.0E-01	1.4E-01	1.4E-01	1.1E-01	1.4E-01	1.0E-01
$NaHCO_3$	2.6E-02	4.4E-02	1.4E-02	2.9E-02	2.6E-02	2.6E-02	4.2E-03		2.6E-02	1.3E-02	2.7E-02	
NaH_2PO_4	1.0E-03	9.1E-04		4.5E-04		1.0E-03				4.2E-03	5.7E-04	
Na_2HPO_4			1.0E-03	5.0E-04			5.6E-03	4.0E-04	1.6E-03		5.0E-04	2.1E-03

(*Continued overleaf*)

TABLE 9.3. Frequently Used Media (*Continued*)

Component	MEM	DMEM	F12	DMEM/ F12	αMEM	CMRL 1066	RPMI 1640	M199	L15	McCoy's 5A	Fischer	MB 752/1
Trace elements												
$CuSO_4 \cdot 5H_2O$			1.6E-08	7.8E-09								
$Fe(NO_3)_3 \cdot 9H_2O$		2.5E-07		1.2E-07								
$FeSO_4 \cdot 7H_2O$			3.0E-06	1.5E-06								
$ZnSO_4 \cdot 7H_2O$			3.0E-06	1.5E-06								
Bases, nucleosides, etc.												
Adenine SO_4								5.4E-05				
Adenosine					3.7E-05							
AMP								5.8E-07				
ATP								1.8E-05				
Cytidine					4.1E-05							
Deoxyadenosine					4.0E-05	4.0E-05						
Deoxycytidine					4.2E-05	3.8E-05						
Deoxyguanosine					3.7E-05	3.7E-05						
2-Deoxyribose								3.7E-06				
DPN						9.5E-06						
FAD						1.2E-06						
Glucuronate, Na						1.9E-05						
Guanine								1.6E-06				
Guanosine					3.5E-05							
Hypoxanthine			3.0E-05	1.5E-05				2.2E-06				
5-Me-deoxycytidine						4.1E-07						
D-Ribose								3.3E-06				
Thymidine			3.0E-06	1.5E-06	4.1E-05	4.1E-05						
Thymine								2.4E-06				
TPN						1.3E-06						
Uracil								2.7E-06				
Uridine					4.1E-05							
UTP						1.8E-06						
Xanthine								2.0E-06				
Energy metabolism												
Cocarboxylase						2.2E-06						
Coenzyme A						3.3E-06						
D-galactose									5.0E-02			
D-glucose	5.6E-03	2.5E-02	1.0E-02	1.8E-02	5.6E-03	5.6E-03	1.1E-02	5.6E-03		1.7E-02	5.6E-03	2.8E-02
Sodium acetate						6.1E-04		4.5E-04				
Sodium pyruvate		1.0E-03	1.0E-03	1.0E-03	1.0E-03				5.0E-03			
Lipids and precursors												
Cholesterol						5.2E-07		5.2E-07				
Ethanol (solvent)						3.5E-04						
Linoleic acid		3.0E-07		1.5E-07								8.9E-05
Lipoic acid			1.0E-06	5.1E-07	9.7E-07							
Tween 80						1.8E-05		1.8E-05				
Other components												
Peptone, mg/mL										0.6		
Phenol red	2.7E-05	4.0E-05	3.2E-05	3.6E-05	2.9E-05	5.3E-05	1.3E-05	4.5E-05	2.7E-05	2.9E-05	1.3E-05	2.7E-05
Putrescine			1.0E-06	5.0E-07								
Gas phase												
CO_2	5%	10%	2%	7%	5%	5%	5%	5%	Air	5%	2%	5%

All concentrations are molar, and computer-style notation is used (e.g., 3.0E-2 = 3.0×10^{-2} = 30 mM). Molecular weights are given for root compounds; although some recipes use salts or hydrated forms, molarities will, of course, remain the same. *Synonyms and abbreviations:* AMP, adenosine monophosphate; ATP, adenosine triphosphate; biotin = vitamin H; calciferol = vitamin D_2; FAD, flavine adenine dinucleotide; lipoic acid = thioctic acid; menadione = vitamin K_3; *myo*-inositol; = L-inostitol; nicotinamide = niacinamide; nicotinic acid = niacin; pyridoxine HCl = vitamin B_6; thiamin = vitamin B_1; α-tocopherol = vitamin E; retinol = vitamin A_1; TPN, triphosphopyridine nucleotide; UTP, uridine triphosphate; vitamin B_{12} = cobalamin. See text for references.

Design de um Incubador

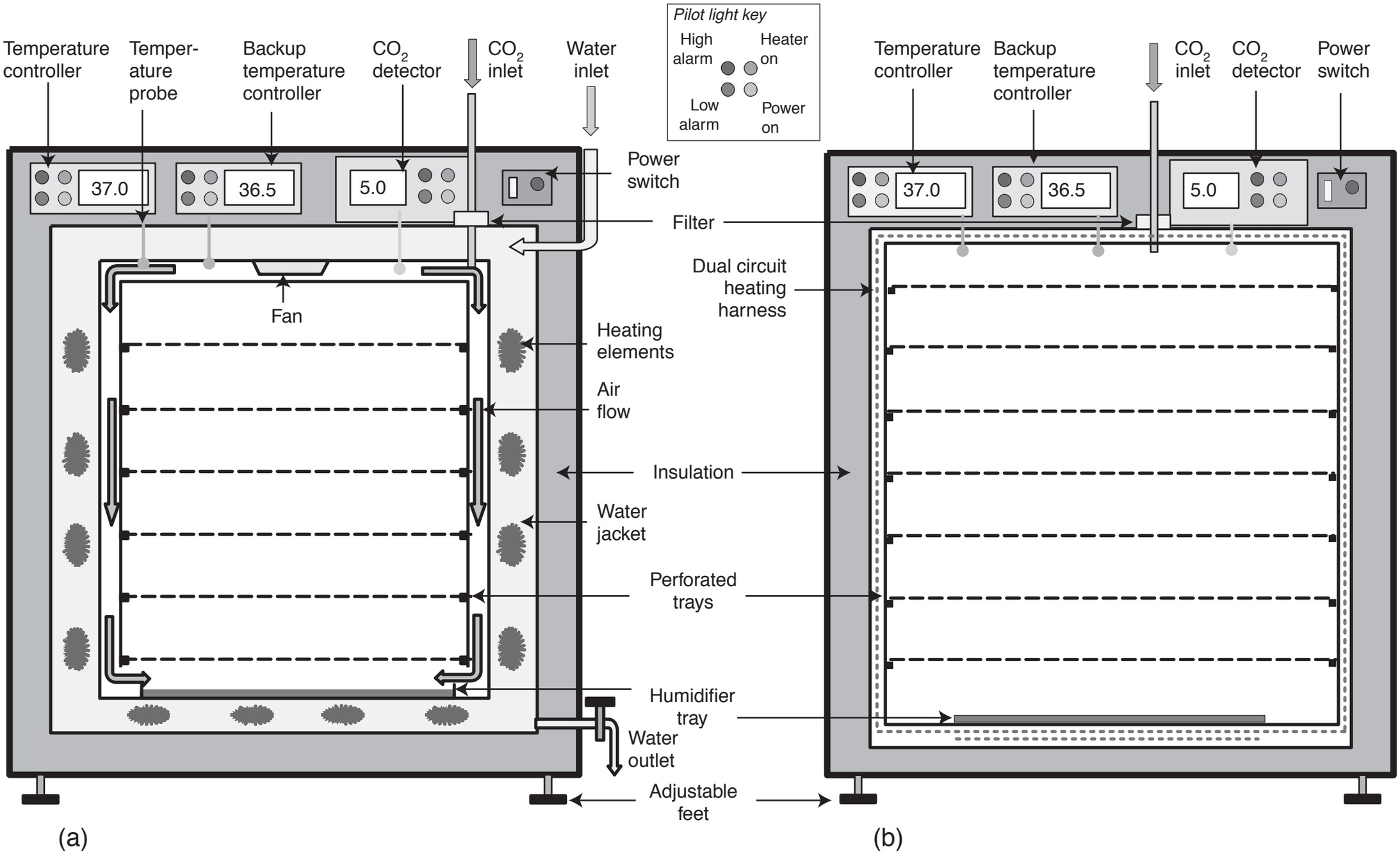


Fig. 5.16. CO_2 Incubator Design. Front view of control panel and section of chamber of two stylized humid CO_2 incubators. (a) Water-jacketed with circulating fan. (b) Dry-walled with no circulating fan (not representative of any particular makes).