

Table S1 Chemically defined medium (CDM) by Otto *et al.*¹

Substance		Concentration
Glucose	$C_6H_{12}O_6$	10 or 40 $g \cdot L^{-1}$
Dipotassium phosphate	K_2HPO_4	2.5 $g \cdot L^{-1}$
Monopotassium phosphate	KH_2PO_4	3 $g \cdot L^{-1}$
Triammonium citrate	$C_6H_{17}N_3O_7$	0.6 $g \cdot L^{-1}$
Sodium acetate	$C_2H_3NaO_2$	1 $g \cdot L^{-1}$
Cysteine hydrochloride	$C_3H_8ClNO_2S$	0.25 $g \cdot L^{-1}$
Salt-free, vitamin-free casein hydrolysate		5 $g \cdot L^{-1}$
100x Vitamin solution		10 $mL \cdot L^{-1}$
100x Metal solution		10 $mL \cdot L^{-1}$
100x Nucleic acid bases solution		10 $mL \cdot L^{-1}$

Vitamin solution for the CDM by Otto *et al.*

Substance		[$mg \cdot L^{-1}$]
Pyridoxine hydrochloride	$C_8H_{12}ClNO_3$	200
Nicotinic acid	$C_6H_5NO_2$	100
Thiamin hydrochloride	$C_{12}H_{18}Cl_2N_4OS$	100
Riboflavin	$C_{17}H_{20}N_4O_6$	100
Calcium D-pantothenate	$C_{18}H_{32}CaN_2O_{10}$	100
Sodium p-aminobenzoate	$C_7H_6NNaO_2$	1000
D-biotin	$C_{10}H_{16}N_2O_3S$	1000
Folic acid	$C_{19}H_{19}N_7O_6$	100
Vitamin B12	$C_{63}H_{88}CoN_{14}O_{14}P$	100
Orotic acid	$C_5H_4N_2O_4$	500
2-deoxythymidine	$C_{10}H_{14}N_2O_5$	500
Inosine	$C_{10}H_{12}N_4O_5$	500
DL-6,8-thioctic acid	$C_8H_{14}O_2S_2$	250
Pyridoxamine hydrochloride	$C_8H_{13}ClN_2O_2$	500

Metal solution for the CDM by Otto *et al.*

Substance		[$mg \cdot L^{-1}$]
Magnesium chloride hexahydrate	$MgCl_2 \cdot 6H_2O$	20
Calcium chloride dihydrate	$CaCl_2 \cdot 2H_2O$	5
Iron chloride tetrahydrate	$FeCl_2 \cdot 4H_2O$	0.5
Zinc sulfate heptahydrate	$ZnSO_4 \cdot 7H_2O$	0.5
Cobalt chloride hexahydrate	$CoCl_2 \cdot 6H_2O$	0.25

Nucleic acid solution for the CDM by Otto *et al.*

Substance		per 10 mL 0.1 M NaOH [g]
Adenine	$C_5H_5N_5$	10
Uracil	$C_4H_4N_2O_2$	10
Xanthine	$C_5H_4N_4O_2$	10
Guanine	$C_5H_5N_5O$	10

Addition of amino acids in the CDM by Poolman and Konings

Substance		Concentration [mg·L ⁻¹]
L-Alanine	C ₃ H ₇ NO ₂	240
L-Arginine	C ₆ H ₁₄ N ₄ O ₂	125
L-Asparagine	C ₄ H ₈ N ₂ O ₃	350
L-Glutamine	C ₅ H ₁₀ N ₂ O ₃	390
Glycine	C ₂ H ₅ NO ₂	175
L-Histidine	C ₆ H ₉ N ₃ O ₂	150
L-Isoleucine	C ₆ H ₁₃ NO ₂	210
L-Leucine	C ₆ H ₁₃ NO ₂	475
L-Lysine	C ₆ H ₁₄ N ₂ O ₂	440
L-Methionine	C ₅ H ₁₁ NO ₂ S	125
L-Phenylalanine	C ₉ H ₁₁ NO ₂	275
L-Proline	C ₅ H ₉ NO ₂	675
L-Serine	C ₃ H ₇ NO ₃	340
L-Threonine	C ₄ H ₉ NO ₃	225
L-Tryptophan	C ₁₁ H ₁₂ N ₂ O ₂	50
L-Tyrosine	C ₉ H ₁₁ NO ₃	200
L-Valine	C ₅ H ₁₁ NO ₂	325

Table S2 CDMPC medium²

Price *et al.* developed a chemically defined medium for prolonged cultivation (CDMPC) of *L. lactis*. The amino acid, metal, vitamin, and alkaline solutions were prepared as stock solutions. After the addition of all components and stock solutions, the medium was sterilized via filtration. Detailed instructions for the preparation are provided by the authors.

Composition of the CDMPC

Substance		Concentration
Glucose	C ₆ H ₁₂ O ₆	10 or 40 g·L ⁻¹
Potassium phosphate	KH ₂ PO ₄	2.75 g·L ⁻¹
Sodium Chloride	NaCl	2.90 g·L ⁻¹
Sodium phosphate	NaH ₂ PO ₄	2.85 g·L ⁻¹
10x amino acid solution		100 mL·L ⁻¹
100x metal solution		10 mL·L ⁻¹
100x vitamin solution		10 mL·L ⁻¹
100x alkaline solution		10 mL·L ⁻¹

10x amino acid solution for the CDMPC

Substance		Concentration [g·L ⁻¹]
L-Alanine	C ₃ H ₇ NO ₂	1.3
L-Arginine	C ₆ H ₁₄ N ₄ O ₂	2.44
L-Asparagine	C ₄ H ₈ N ₂ O ₃	0.8
L-Aspartic acid	C ₄ H ₇ NO ₄	1.37
L-Cysteine hydrochloride monohydrate	C ₃ H ₇ NO ₂ S HCl H ₂ O	0.61
L-Glutamic acid	C ₅ H ₉ NO ₄	0.97

L-Glutamine	$C_5H_{10}N_2O_3$	0.96
Glycine	$C_2H_5NO_2$	0.29
L-Histidine	$C_6H_9N_3O_2$	0.24
L-Isoleucine	$C_6H_{13}NO_2$	0.82
L-Leucine	$C_6H_{13}NO_2$	1.17
L-Lysine monohydrochloride	$C_6H_{14}N_2O_2$ HCl	1.87
L-Methionine	$C_5H_{11}NO_2S$	0.38
L-Phenylalanine	$C_9H_{11}NO_2$	0.64
L-Proline	$C_5H_9NO_2$	4.12
L-Serine	$C_3H_7NO_3$	1.72
L-Threonine	$C_4H_9NO_3$	0.68
L-Tryptophan	$C_{11}H_{12}N_2O_2$	0.36
L-Valine	$C_5H_{11}NO_2$	0.86

100x metal solution for the CDMPC

Substance	Molecular formula	Concentration	[mg·L ⁻¹]
Ammonium molybdate tetrahydrate	$(NH_4)_6Mo_7O_{24} \cdot 4H_2O$		30
Calcium chloride dihydrate	$CaCl_2 \cdot 2H_2O$		300
Cobalt sulfate heptahydrate	$CoSO_4 \cdot 7H_2O$		30
Copper sulfate pentahydrate	$CuSO_4 \cdot 5H_2O$		30
Iron chloride tetrahydrate	$FeCl_2 \cdot 4H_2O$		400
Magnesium chloride hexahydrate	$MgCl_2 \cdot 6H_2O$		20,000
Manganese chloride tetrahydrate	$MnCl_2 \cdot 4H_2O$		400
Zinc sulfate heptahydrate	$ZnSO_4 \cdot 7H_2O$		30

100x vitamin solution for the CDMPC

Substance			[mg·L ⁻¹]
α -Lipoic acid	$C_8H_{14}O_2S_2$		200
D-Pantothenic acid hemicalcium salt	$C_9H_{16}NO_5$ 1/2 Ca		50
Nicotinic acid	$C_6H_5NO_2$		100
Pyridoxal hydrochloride	$C_8H_9NO_3$ HCl		100
Pyridoxine hydrochloride	$C_8H_{11}NO_3$ HCl		100
Thiamine hydrochloride	$C_{12}H_{17}ClN_4OS$ HCl		100

100x alkaline solution for the CDMPC

Substance			[mg·L ⁻¹]
Biotin	$C_{10}H_{16}N_2O_3S$		10
L-Tyrosine	$C_9H_{11}NO_3$		5000

Table S3 Resting cell buffer

Resting cell experiments were conducted in a buffer solution. The phosphate buffer, trace elements, and magnesium solution were prepared as individual stock solutions in 100x concentration. The glucose solution was prepared as a 500 g·L⁻¹ stock solution. All stock solutions were autoclaved except for the trace element solution which was sterile filtered. For the preparation of the washing buffer, used for the washing step during resting cell experiments, glucose was omitted.

Composition of the resting cell buffer

Phosphate buffer

Dipotassium phosphate	K_2HPO_4	$3.88 \text{ g}\cdot\text{L}^{-1}$
Monosodium phosphate	NaH_2PO_4	$1.63 \text{ g}\cdot\text{L}^{-1}$
Magnesium sulfate	$MgSO_4$	$0.25 \text{ g}\cdot\text{L}^{-1}$

Trace elements

EDTA	$C_{10}H_{16}N_2O_8$	$10 \text{ mg}\cdot\text{L}^{-1}$
Magnesium chloride hexahydrate	$MgCl_2 \cdot 6H_2O$	$0.10 \text{ g}\cdot\text{L}^{-1}$
Zinc sulfate heptahydrate	$ZnSO_4 \cdot 7H_2O$	$2 \text{ mg}\cdot\text{L}^{-1}$
Calcium chloride dihydrate	$CaCl_2 \cdot 2H_2O$	$1 \text{ mg}\cdot\text{L}^{-1}$
Iron sulfate heptahydrate	$FeSO_4 \cdot 7H_2O$	$5 \text{ mg}\cdot\text{L}^{-1}$
Sodium molybdate dihydrate	$Na_2MoO_4 \cdot 2H_2O$	$0.2 \text{ mg}\cdot\text{L}^{-1}$
Copper sulfate pentahydrate	$CuSO_4 \cdot 5H_2O$	$0.2 \text{ mg}\cdot\text{L}^{-1}$
Cobalt chloride hexahydrate	$CoCl_2 \cdot 6H_2O$	$0.4 \text{ mg}\cdot\text{L}^{-1}$
Manganese chloride dihydrate	$MnCl_2 \cdot 2H_2O$	$1 \text{ mg}\cdot\text{L}^{-1}$
Carbon source glucose	$C_6H_{12}O_6$	$40 \text{ g}\cdot\text{L}^{-1}$

Table S4 Variation of filling volume in 500 mL shake flasks to vary the oxygen transfer for cultivations of *L. lactis* VJ017 and the corresponding process parameters. ^aThe oxygen transfer rate was calculated according to Meier *et al*³.

Filling volume [mL]	Glucose concentration [$\text{g}\cdot\text{L}^{-1}$]	OTR [$\text{mmol}\cdot\text{L}^{-1}\cdot\text{h}^{-1}$] ^a	Acetoin titer [$\text{g}\cdot\text{L}^{-1}$]	Acetoin yield [$\text{g}_{\text{acetoin}}^{-1}\cdot\text{g}_{\text{glucose}}^{-1}$]	Specific acetoin yield [$\text{g}_{\text{acetoin}}^{-1}\cdot\text{g}_{\text{CDW}}^{-1}$]
50	40	28.62	13.8 ± 0.1	0.35 ± 0.0	2.26 ± 0.03
25	40	47.80	16.5 ± 0.3	0.41 ± 0.01	2.97 ± 0.16
25	10	47.80	4.8 ± 0.1	0.48 ± 0.00	1.85 ± 0.13

Table S5 Acetoin concentration in the respective media and applied current densities for electrochemical conversion of acetoin to 2-butanone.

Medium	Concentration [$\text{g}\cdot\text{L}^{-1}$]	Current Density [$\text{mA}\cdot\text{cm}^{-2}$]
M17	6	10
HS	7	10
LB	8	10
YEP	14	25
M17	16	25
Resting cell buffer	19	25

1. R. Otto, B. ten Brink, H. Veldkamp and W. N. Konings, *FEMS Microbiology Letters*, 1983, **16**, 69-74.
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3. K. Meier, W. Klöckner, B. Bonhage, E. Antonov, L. Regestein and J. Büchs, *Biochemical Engineering Journal*, 2016, **109**, 228-235.