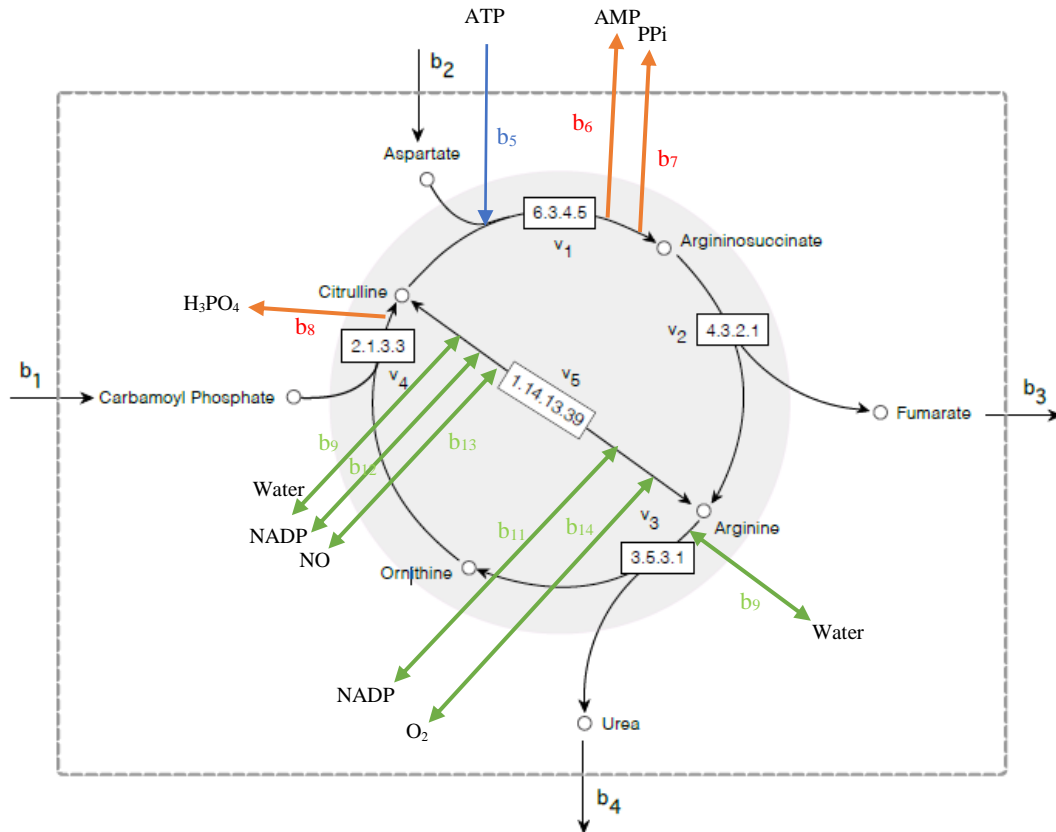


## PS 3 Solution



**Figure 1:** Schematic of the Urea cycle.

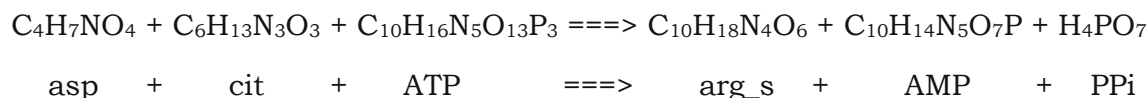
Solution 1 (a)

▪ Metabolite ID

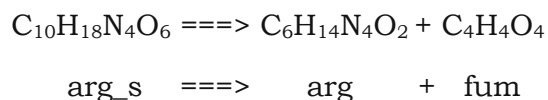
#	Metabolite	Symbol	#	Metabolite	Symbol
M.1	Arginine (arg)	C <sub>6</sub> H <sub>14</sub> N <sub>4</sub> O <sub>2</sub>	M.10	ATP	C <sub>10</sub> H <sub>16</sub> N <sub>5</sub> O <sub>13</sub> P <sub>3</sub>
M.2	Ornithine (orn)	C <sub>5</sub> H <sub>12</sub> N <sub>2</sub> O <sub>2</sub>	M.11	AMP	C <sub>10</sub> H <sub>14</sub> N <sub>5</sub> O <sub>7</sub> P
M.3	Urea (u)	CH <sub>4</sub> N <sub>2</sub> O	M.12	Diphosphate (PPi)	H <sub>4</sub> PO <sub>7</sub>
M.4	Carbamoyl Phosphate (camp)	CH <sub>4</sub> NO <sub>5</sub> P	M.13	Orthophosphate	H <sub>3</sub> PO <sub>4</sub>
M.5	L-aspartate (asp)	C <sub>4</sub> H <sub>7</sub> NO <sub>4</sub>	M.14	NADPH	C <sub>21</sub> H <sub>30</sub> N <sub>7</sub> O <sub>17</sub> P <sub>3</sub>
M.6	Citrulline (cit)	C <sub>6</sub> H <sub>13</sub> N <sub>3</sub> O <sub>3</sub>	M.15	NADP	C <sub>21</sub> H <sub>29</sub> N <sub>7</sub> O <sub>17</sub> P <sub>3</sub>
M.7	Arginine succinate (arg_s)	C <sub>10</sub> H <sub>18</sub> N <sub>4</sub> O <sub>6</sub>	M.16	Oxygen	O <sub>2</sub>
M.8	Fumarate (fum)	C <sub>4</sub> H <sub>4</sub> O <sub>4</sub>	M.17	Nitric Oxide	NO
M.9	Water	H <sub>2</sub> O	M.18	H <sup>+</sup>	H

▪ Reactions involved in the urea cycle (Source: [KEGG](#))

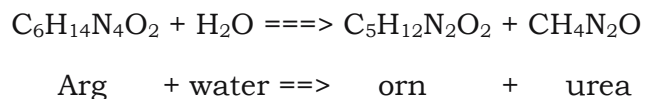
**E.C. 6.3.4.5**



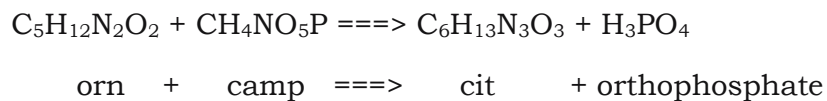
**E.C. 4.3.2.1**



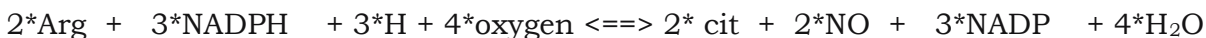
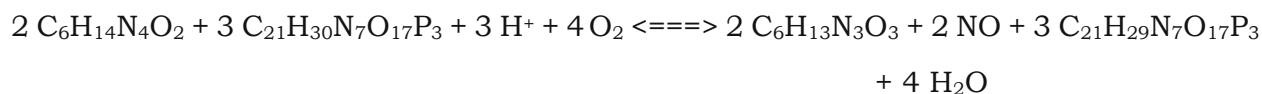
**E.C. 3.5.3.1**



**E.C. 2.1.3.3**



### E.C. 1.14.13.39



#### Reaction ID

#	Reaction	#	Reaction
v <sub>1</sub>	<i>e.c.6.3.4.5</i> asp-->arg_s	b <sub>5</sub>	[ ]-->ATP
v <sub>2</sub>	<i>e.c.4.3.2.1</i> arg_s-->arg	b <sub>6</sub>	AMP-->[ ]
v <sub>3</sub>	<i>e.c.3.5.3.1</i> arg-->orn	b <sub>7</sub>	PPi-->[ ]
v <sub>4</sub>	<i>e.c.2.1.3.3</i> orn-->cit	b <sub>8</sub>	H <sub>3</sub> PO <sub>4</sub> -->[ ]
v <sub>5</sub>	<i>e.c.1.14.13.39</i> arg-->cit	b <sub>9</sub>	[ ]<-->H <sub>2</sub> O
v <sub>6</sub>	rev. v <sub>5</sub> <i>e.c.1.14.13.39</i> cit-->arg	b <sub>10</sub>	NADPH<-->[ ]
b <sub>1</sub>	[ ]-->camp	b <sub>11</sub>	H<-->[ ]
b <sub>2</sub>	[ ]-->asp	b <sub>12</sub>	NADP<-->[ ]
b <sub>3</sub>	fum-->[ ]	b <sub>13</sub>	NO<-->[ ]
b <sub>4</sub>	u-->[ ]	b <sub>14</sub>	O <sub>2</sub> <-->[ ]

#### (answer) Stoichiometric Matrix, S

	v <sub>1</sub>	v <sub>2</sub>	v <sub>3</sub>	v <sub>4</sub>	v <sub>5</sub>	v <sub>6</sub>	b <sub>1</sub>	b <sub>2</sub>	b <sub>3</sub>	b <sub>4</sub>	b <sub>5</sub>	b <sub>6</sub>	b <sub>7</sub>	b <sub>8</sub>	b <sub>9</sub>	b <sub>10</sub>	b <sub>11</sub>	b <sub>12</sub>	b <sub>13</sub>	b <sub>14</sub>
M.1	0	1	-1	0	-2	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M.2	0	0	1	-1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M.3	0	0	1	0	0	0	0	0	0	-1	0	0	0	0	0	0	0	0	0	0
M.4	0	0	0	-1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
M.5	-1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
M.6	-1	0	0	1	2	-2	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M.7	1	-1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
M.8	0	1	0	0	0	0	0	0	-1	0	0	0	0	0	0	0	0	0	0	0
M.9	0	0	-1	0	4	-4	0	0	0	0	0	0	0	0	1	0	0	0	0	0
M.10	-1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
M.11	1	0	0	0	0	0	0	0	0	0	0	-1	0	0	0	0	0	0	0	0
M.12	1	0	0	0	0	0	0	0	0	0	0	0	-1	0	0	0	0	0	0	0
M.13	0	0	0	1	0	0	0	0	0	0	0	0	0	-1	0	0	0	0	0	0
M.14	0	0	0	0	-3	3	0	0	0	0	0	0	0	0	0	-1	0	0	0	0
M.15	0	0	0	0	3	-3	0	0	0	0	0	0	0	0	0	0	0	1	0	0
M.16	0	0	0	0	-4	4	0	0	0	0	0	0	0	0	0	0	0	0	0	-1
M.17	0	0	0	0	2	-2	0	0	0	0	0	0	0	0	0	0	0	0	1	0
M.18	0	0	0	0	-3	3	0	0	0	0	0	0	0	0	0	0	-1	0	0	0

(Convention for v<sub>5</sub> reversibility has been assumed to be: citrulline ==> arginine)

### Solution 1 (b)

#### Atom Matrix, A

	M.1	M.2	M.3	M.4	M.5	M.6	M.7	M.8	M.9	M.10	M.11	M.12	M.13	M.14	M.15	M.16	M.17	M.18
C	6	5	1	1	4	6	10	4	0	10	10	0	0	21	21	0	0	0
H	14	12	4	4	7	13	18	4	2	16	14	4	3	30	29	0	0	1
N	4	2	2	1	1	3	4	0	0	5	5	0	0	7	7	0	1	0
O	2	2	1	5	4	3	6	4	1	13	7	7	4	17	17	2	1	0
P	0	0	0	1	0	0	0	0	0	3	1	2	1	3	3	0	0	0

I have used the atom matrix for this part and the **epsilon** = **A\*S** matrix can be generated by running the ("**S\_and\_epsilon\_generator.jl**") file.

- in julia console run `include("a,b S_and_epsilon_generator.jl")`

This will generate "atom\_matrix.csv" and "epsilon.txt" files.

If the urea cycle reconstruction is elementally balanced, all components of the epsilon matrix should be zero, which is true in this case. (view "epsilon.txt")

### Solution 1 (c)

The code for solving the FBA problem is given in "c run.jl"

- Upper bounds for the fluxes were calculated using:

$$v_{max,i} = k_{cat,i} \times E \times \prod_{i=1}^m \frac{C_i}{K_m + C_i}$$

- $K_m$  values and metabolite concentration,  $C_i$  were obtained from Park et al Nat Chem Biol 12:482-9, 2016 and used to calculate:

For these values, the following order of preference was followed:

*Homo sapiens > Mus musculus > other organism*

Data for *S. cerevisiae* was used for the enzyme E.C.2.1.3.3

*Note:* I have assumed the metabolites involved in reaction  $v_5$  can be exchanged through the boundary. Deactivate lines 25, 26 and activate lines 29 and 30 to see fluxes with **no boundary exchange of reaction  $v_5$  metabolites** (in this case flux  $v_5 = v_6 = 0.0$  mmol/gDW.hr to maximize urea)

- in julia console run `include("c run.jl")`

**(answer)** Optimum fluxes for maximum urea production

Flux ID	Reaction	LB	UB	Optimum value (mmol/gdW.hr)
v <sub>1</sub>	<i>e.c.6.3.4.5</i> asp-->arg_s	0	V <sub>max1</sub>	1.242
v <sub>2</sub>	<i>e.c.4.3.2.1</i> arg_s-->arg	0	V <sub>max2</sub>	1.242
v <sub>3</sub>	<i>e.c.3.5.3.1</i> arg-->orn	0	V <sub>max3</sub>	1.271095
v <sub>4</sub>	<i>e.c.2.1.3.3</i> orn-->cit	0	V <sub>max4</sub>	1.271095
v <sub>5</sub>	<i>e.c.1.14.13.39</i> arg-->cit	0	V <sub>max5</sub>	0
v <sub>6</sub>	rev. v <sub>5</sub> <i>e.c.1.14.13.39</i> cit-->arg	0	V <sub>max6</sub>	0.014548
b <sub>1</sub>	[ ]-->camp	0	10	1.271095
b <sub>2</sub>	[ ]-->asp	0	10	1.242
b <sub>3</sub>	fum-->[ ]	0	10	1.242
<b>b<sub>4</sub></b>	<b>u--&gt;[ ]</b>	<b>0</b>	<b>10</b>	<b>1.271095</b>
b <sub>5</sub>	[ ]-->ATP	0	10	1.242
b <sub>6</sub>	AMP-->[ ]	0	10	1.242
b <sub>7</sub>	PPi-->[ ]	0	10	1.242
b <sub>8</sub>	H <sub>3</sub> PO <sub>4</sub> -->[ ]	0	10	1.271095
b <sub>9</sub>	[ ]<-->H <sub>2</sub> O	-10	10	1.329286
b <sub>10</sub>	NADPH<-->[ ]	-10	10	0.043643
b <sub>11</sub>	H<-->[ ]	-10	10	0.043643
b <sub>12</sub>	NADP<-->[ ]	-10	10	0.043643
b <sub>13</sub>	NO<-->[ ]	-10	10	0.029095
b <sub>14</sub>	O <sub>2</sub> <-->[ ]	-10	10	0.05819