Part A

The reaction cycle given in the problem set statement is not the full reaction. There are other substrates missing in the Urea cycle. To construct a full stoichiometric matrix S for this cycle, KEGG (Arginine biosynthesis in human) was used as the tool of knowing the complete reaction. The following stoichiometric matrix is 18×20, representing 18 metabolites and 20 flux needed.

$$\begin{array}{c}
\begin{bmatrix}
0 & 0 & 0 & -1 & 0 & 0 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
-1 & 0 & 0 & 1 & -1 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
-1 & 0 & 0 & 0 & 0 & 0 & 0 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
1 & -1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
0 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & -1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
0 & 1 & -1 & 0 & 1 & -1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
0 & 0 & 1 & -1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
-1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
0 & 0 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & -1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
0 & 0 & 0 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & -1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & -1 & 0 & 0 & 0 & 0 & 0 & 0 \\
1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & -1 & 0 & 0 & 0 & 0 & 0 \\
0 & 0 & 0 & 0 & -1 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & -1 & 0 & 0 & 0 & 0 \\
0 & 0 & 0 & 0 & 2 & -2 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & -1 & 0 & 0 & 0 \\
0 & 0 & 0 & 0 & 1.5 & -1.5 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 1 & 0 & 0 \\
0 & 0 & 0 & 0 & 1.5 & -1.5 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 1 & 0 \\
0 & 0 & 0 & 0 & -1.5 & 1.5 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & -1 \\
0 & 0 & -1 & 0 & -2 & 2 & 0 & 0 & 0 & 0 & 0 & 1 & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0
\end{bmatrix} \\
\begin{matrix} v1 & v2 & v3 & v4 & v5 & v5 & b1 & b2 & b3 & b4 & b5 & b6 & b7 & b8 & b9 & b10 & b11 & b12 & b13 & b14 \end{matrix}
\end{array}$$

The table below shows how metabolites correspond to row numbers.

Row #	Metabolite	Row #	Metabolite
1	Carbamoyl Phosphate	10	Pi
2	Citrulline	11	AMP
3	Aspartate	12	PPi
4	Arginosuccinate	13	NO
5	Fumarate	14	O2
6	Arginine	15	H^+
7	Ornithine	16	NADPH
8	ATP	17	$NADP^+$
9	Urea	18	H_2O

The fluxes are defined as given in the problem statement. For the reactions not shown in the given graph, I defined them as the following:

b5: ATP

b6: H_2O

b7: phosphate

b8: AMP

b9: Pyrophaphate

b10: Nitric Oxide Sink

b11: O_2

b12: H^+

b13: NADPH

b14: $NADP^+$

Part B

The balance should be written around the metabolites for C, H, N, O, P and S. To test if it is balanced, we want to use the equation A×S=E. If E=0, the atoms are balanced well. If E≠0, there might be some atoms not balanced. Matrix S is the stoichiometric matrix written in part a). Matrix A can be written for each atom as the following:

	Carbamoyl Phosphate	Citrulline	Aspartate	Arginosuccinate	Fumarate	Arginine	Ornithine	ATP	Urea	Pi	AMP	PPi	NO	O2	H+	NADPH	NADP+	H2O
C	1	6	4	10	4	6	5	10	1	0	10	0	0	0	0	21	21	0
H	4	13	7	18	4	14	12	16	4	3	14	4	0	0	1	30	29	2
N	1	3	1	4	0	4	2	5	2	0	5	0	1	0	0	7	7	0
O	5	3	4	6	4	2	2	13	1	4	7	7	1	2	0	17	17	1
P	1	0	0	0	0	0	0	3	0	1	1	2	0	0	0	3	3	0
S	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

The result shows that some elements in some reactions are not balanced. To make them balance, we need to change stoichiometric matrix until each entry of E is 0.

Part C

In part C, it is required to find maximum rate of urea production (b_4) by using FBA method. The constraints obey that

$$0 \leqslant vj \leqslant kcat,j \times E \times \theta \times \Pi \frac{X_j}{K_M + X_i}$$

$$0 \leqslant b_j \leqslant 10 \text{ mmol/gDW-hr}$$

By using the given Julia file flux.jl, we can write a Julia code that utilizes the finding in part a to get the maximum rat of urea production. The final result obtained from Julia programming is 1033.4 umol/gDW/hr.