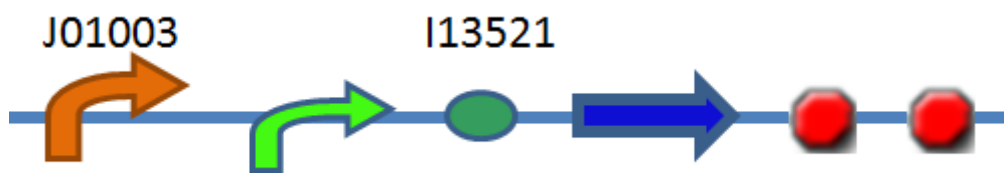


The confirmation experiment of Conjugation

Because this part contains oriT, the R plasmid nic region is where the relaxosome nicks the plasmid and conjugative transfer by R plasmid machinery begins. The function of this kit (K873003) is: when there is a RP4 plasmid, conjugation will be induced to start. We use BL21 which carry the K873003 plasmid as donor bacteria while DH5 α acts as recipient bacteria, and the helper is the HB101 strain who carry the RP4 plasmid. According to the document, DH5 α anti Nalidixic Acid HB101 has resistance over Ampicillin and Kanamycin, and BL21 has Chloramphenicol resistance. If the conjugation happens, the recipient bacterium will not only anti Nalidixic Acid, but also have Chloramphenicol resistance .



Experimental procedure:

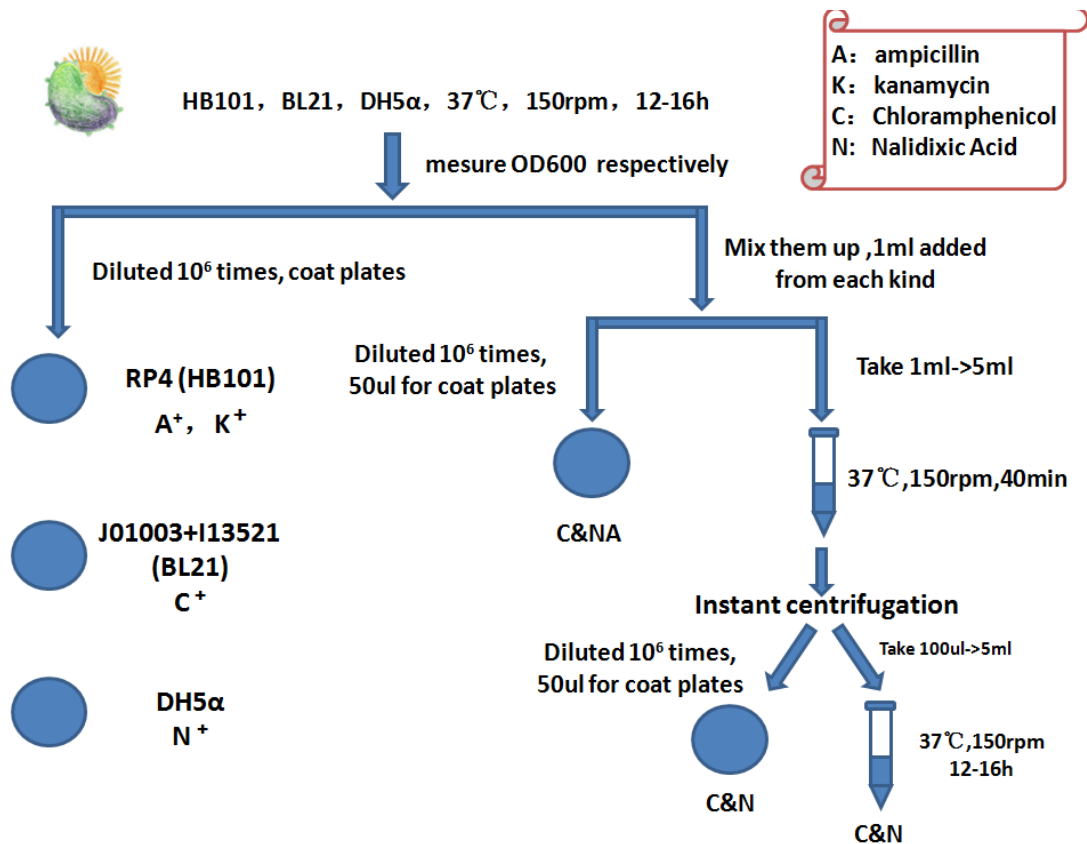


Fig1. The experimental procedure of Sweet gene's confirmation experiment

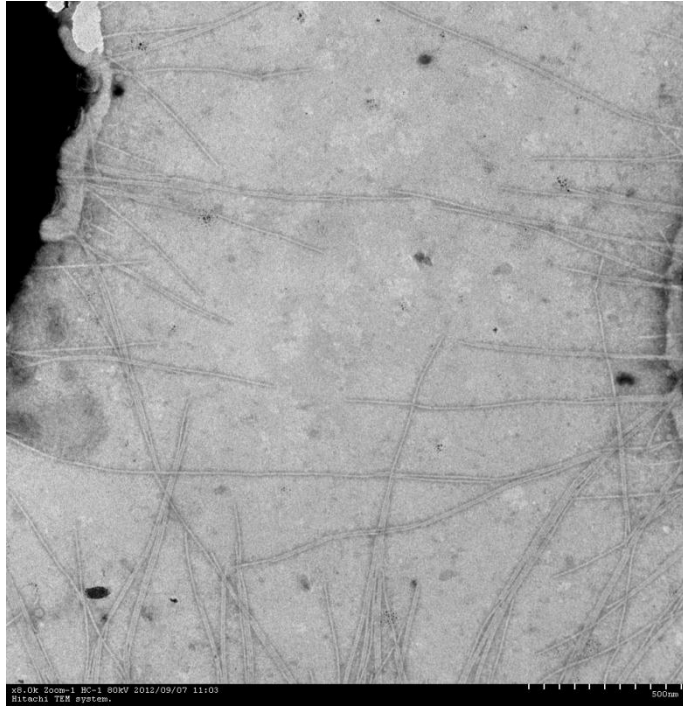


Fig2. The transmission electron microscope image of conjugation

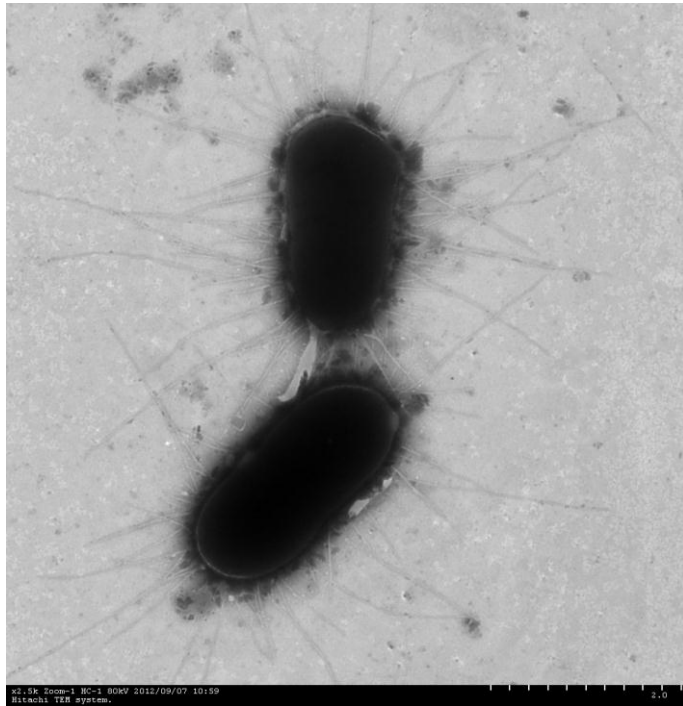


Fig3. The TEM image of conjugation

The data of Sweet gene's confirmation experiment version 1.0

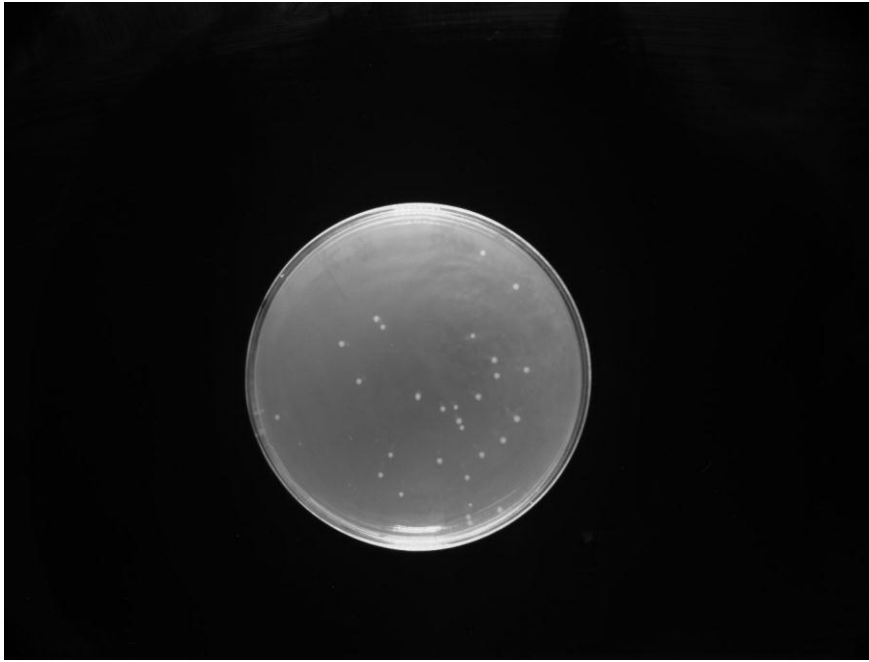


Fig4. The agar Luria-Bertani medium of DH5 α

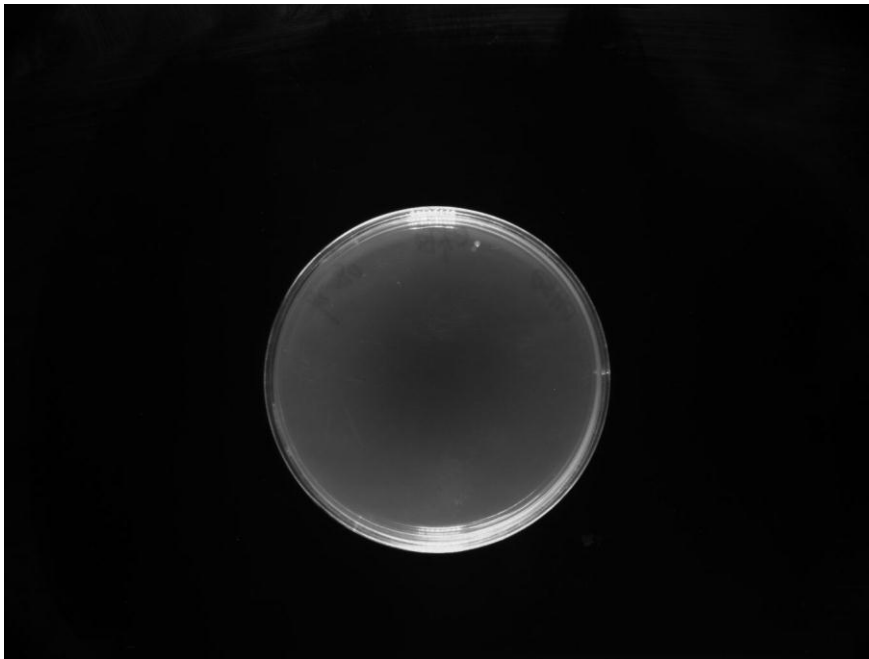


Fig 5. The agar Luria-Bertani medium of DH5 α

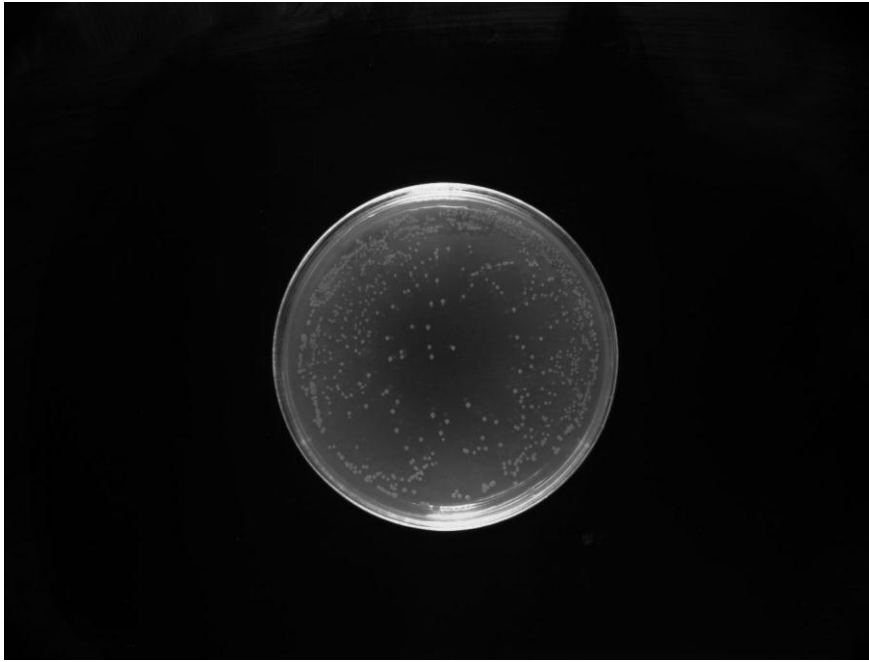


Fig 6. The agar Luria-Bertani medium of BL21

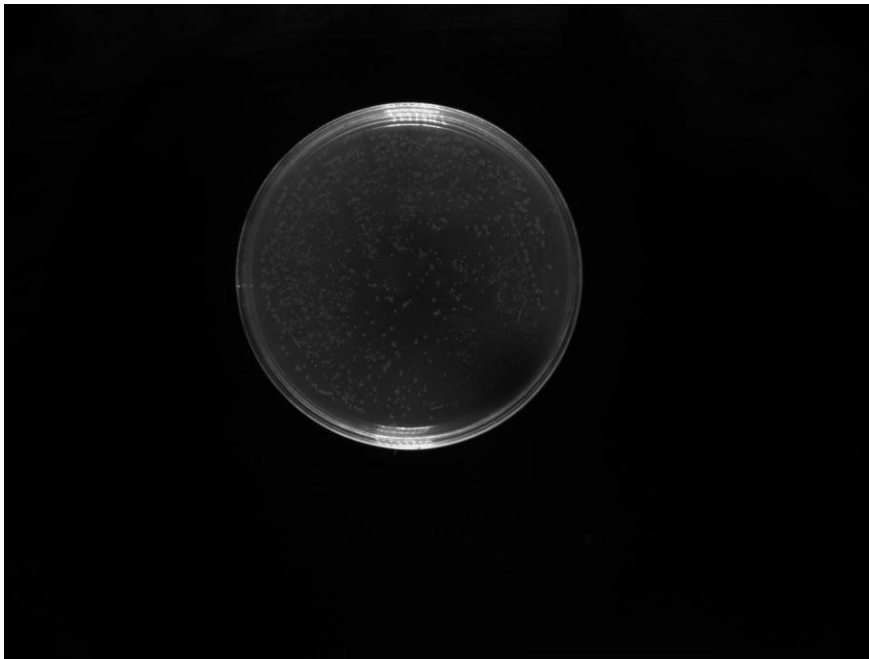


Fig7. The agar Luria-Bertani medium of BL21

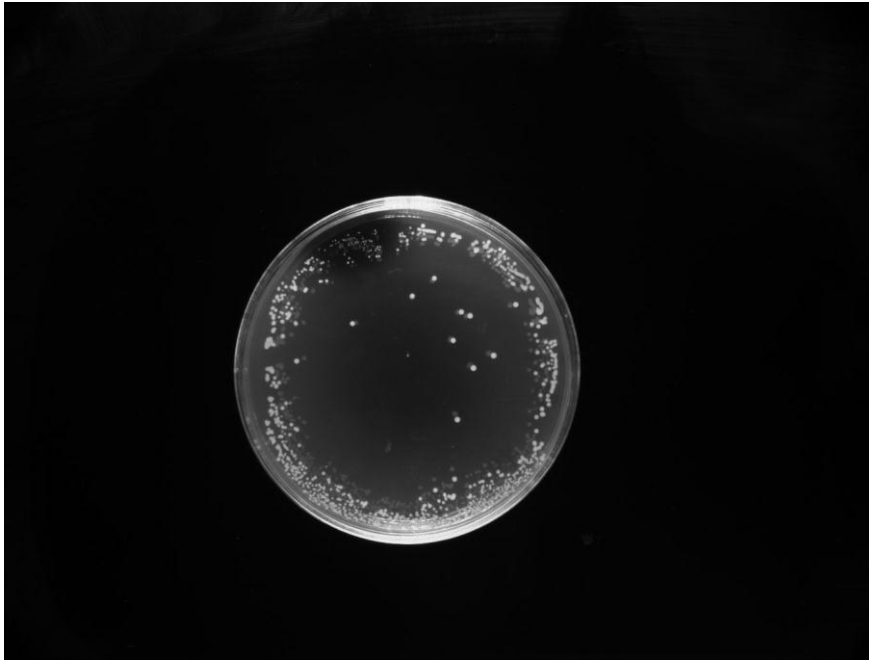


Fig8. The agar Luria-Bertani medium of HB101



Fig9. The agar Luria-Bertani medium of HB101



Fig10. The agar Luria-Bertani medium of conjugation

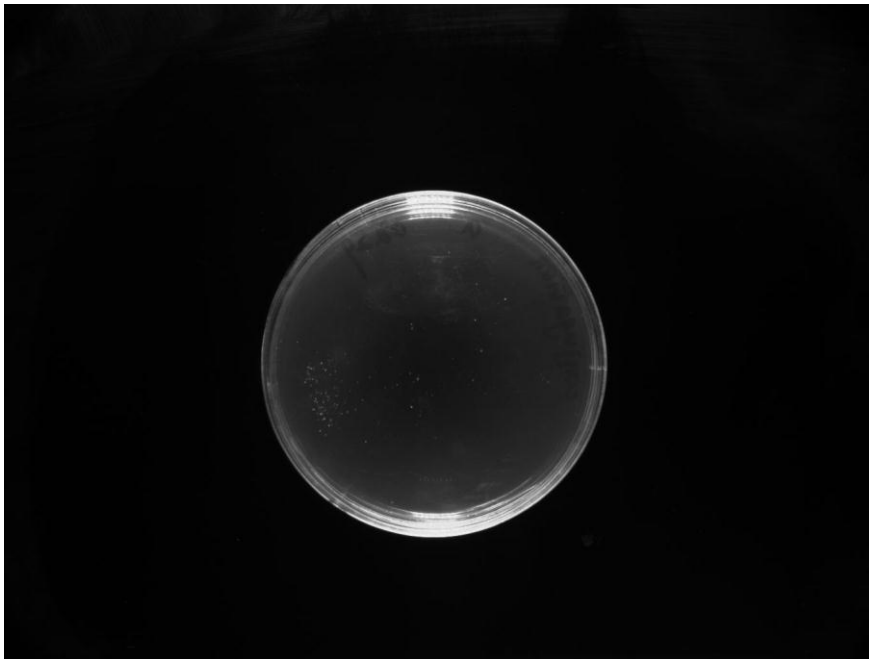


Fig11. The agar Luria-Bertani medium of conjugation

We process the picture by the model of our member Haoruo Jia. The results are as follows:

Cell	Number	Total area	Accuracy class	P1	P2	P3	Level(0-1)
DH5 α	28	846	Nice	800	100	15	0.6
DH5 α	2	162	Nice	800	80	70	0.4

BL21	439	8951	Good	800	80	8	0.32
BL21	765	10421	Acceptable	800	80	3	0.18
HB101	381	8700	Good	800	80	3	0.55
HB101	2255	28262	Good	800	80	3	0.35
Transconjugants	72	648	Good	200	80	5	0.2
Transconjugants	130	1819	Good	1000	80	7	0.2

Thus the efficiency of conjugation is: $\text{Tranconjugants/BL21} = 0.12347$.