

(1) Determination of the relationship between fluorescence value and growth time

1. Purpose of the experiment: the effect of secondary light-involved system with culture time.

2. Experimental equipment

2.1. Reagents: bacterial solution of BL21+T7-dusk-eGFP-pUC57 (10μg/ml IPTG) and BL21+T7-dusk-eGFP-pUC57 for a certain time, M9 liquid medium.

2.2. Instruments: liquid transfer guns and gun heads, alcohol lamp, 96-well-black board, fluorescent enzyme marker, etc.

3. experimental steps

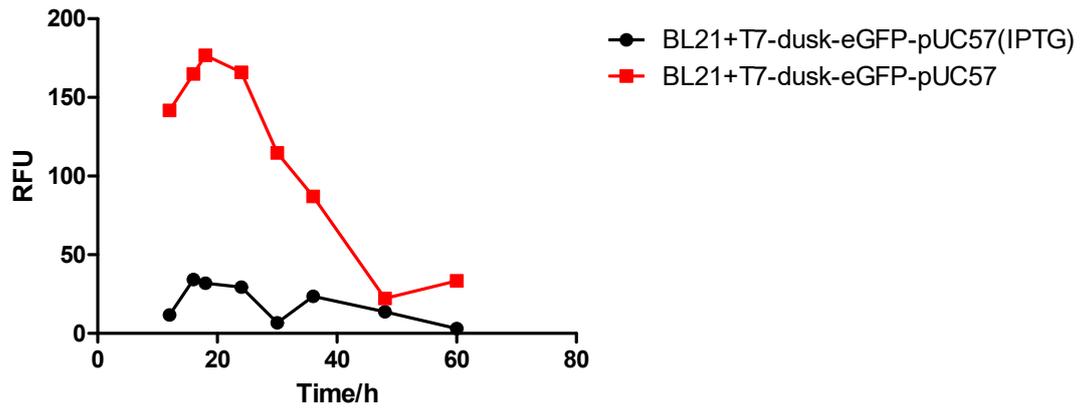
Next to the ignited alcohol lamp, the control group and the experimental group cultured for a certain time were added 100μl to the 96-well-black board according to the table below, and the fluorescence value was detected by fluorescence enzyme labeling instrument (excitation wavelength: 488nm; emission wavelength 516nm).

Number	Bacterial solution	Condition of culture	Time
A	BL21+T7-dusk-eGFP-pUC57(10μg/ml IPTG)	37 °C vibration culture without light	12h、16h、18h、24h、30h、36h、48h、60h
B	BL21+T7-dusk-eGFP-pUC57		

4. Determination results

RFU Time	BS	BL21+T7-dusk-eGFP-pUC57(10μg/ml IPTG)			BL21+T7-dusk-eGFP-pUC57		
		12h	47.584	56.079	58.506	147.183	157.313
16h	68.546	69.989	69.66	196.139	201.209	200.372	
18h	77.026	79.402	78.452	208.68	208.615	208.793	
24h	53.707	51.1	51.302	192.246	195.643	198.226	
30h	45.364	42.75	44.594	120.364	122.75	121.594	
36h	43.581	43.567	42.823	111.008	109.843	110.91	
48h	45.889	47.563	45.981	35.75	35.918	36.242	
60h	34.102	33.638	33.567	36.728	35.699	37.084	

RFU Time	BS	M9 liquid medium		
		12h	38.25	41.464
16h	37.883	38.136	29.321	
18h	47.583	46.05	45.61	
24h	21.375	24.212	22.035	
30h	34.124	41.593	36.583	
36h	17.724	19.625	22.091	
48h	29.148	32.679	36.345	
60h	32.454	31.217	28.284	



5. Analyse

According to the experimental results, the fluorescence value of the secondary light-involved system added with IPTG was always lower than that without IPTG, which may be due to the inappropriate amount of IPTG added in the M9 medium. Therefore, it was decided to re-examine the relationship between the amount of IPTG and the fluorescence value in the M9 medium.