E.coli Curdlan Purification

Overview :

Curdlan molecules may have as many as 12,000 glucose units and are insoluble in water, alcohols and most organic solvents,while it is soluble in dilute alkali (0.25 M NaOH).
We have two protocols for Curdlan purification from *E.coli* based on this properties to be soluble in NaOH solution.

Procedure n°1 :

Reference : Demonstration of Curdlan-type polysaccharide and some other in microorganisms with aniline blue, Itaru Nakanishi, Tokuya Harada and all, J. Gen. App!. Microbiol., 22,1-11(1976)

1) The culture was mixed with an equal volume of 1 N NaOH and then centrifuged at 10,000 rpm for 10min to remove the cells.
2) The resulting supernatant was neutralized by adding 3 N HCl
3) The precipitate formed was collected by centrifugation at 10,000rpm for 10min.
   -It was washed three times with water by centrifugation, and then dehydrated with acetone and dried in vacuo.
4) This precipitated polymer is the curdlan-type polysaccharide.

Procedure n°2 :

Reference : « Exopolymers from curdlan production: incorporation of glucose-related sugars by Agrobacterium sp. strain ATCC 31749. Jin W. Lee & all, Canadian Journal of Microbiology, 1997, Vol. 43, N° 2 : pages 149-156 »

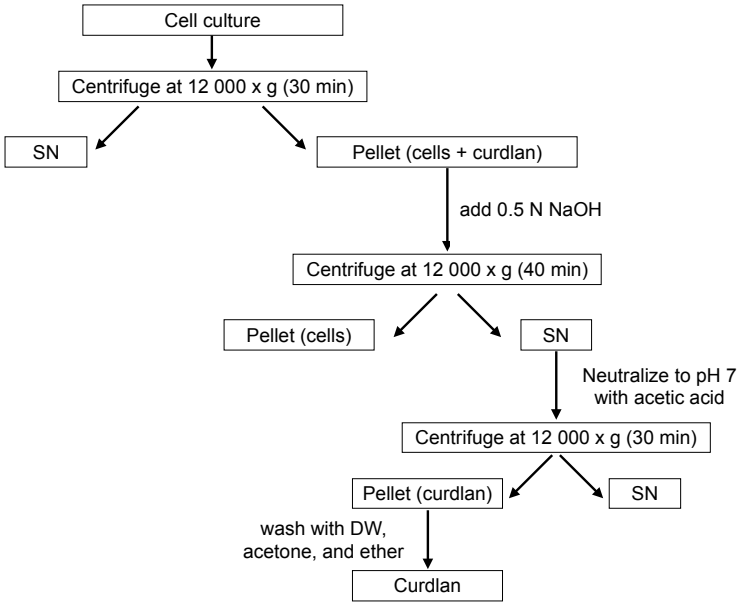

Fig. 1. Flow diagram depicting the purification procedure for curdlan. SN, supernatant.
Adapted from the reference.

1) The culture was centrifuged at 12000 x g for 30 min.
2) The pellet was added to an equivalent volume of 0.5 N sodium hydroxide at 3°C
3) The mixture was stirred for 10 min
4) Left to stand for 3 h at the same temperature.
5) The resulting viscous solution was centrifuged at 12000 x g for 40 min
6) Curdlan in the clear supernatant was precipitated by neutralization with 10% acetic acid
7) Repeatedly washed with DWater, acetone and ether.