HIGH CELL DENSITY CULTIVATION OF PENICILLIUM CHRYSOGENUM：PRODUCTION OF PENICILLIN V IN A FERMENTOR

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ABSTRACT

PENICILLIN IS A KIND OF THE SECONDARY METABOLITES PRODUCED BY PENICLLIUM SP., AND IT POSSESSES ANTIMICROBIAL CAPABILITY. PENICILLIN CAN BE CLASSIFIED INTO THREE CATEGORIES AS NATURAL, BIOSYNTHETIC AND SEMI-BIOSYNTHETIC PENICILLINS ACCORDING TO THEIR BINDING SIDE-CHAINS. PENICILLIN V BELONGS TO THE CATEGORY OF BIOSYNTHETIC PENICILLINS, AND IT IS QUITE STABLE IN ACIDIC ENVIRONMENT, AND CAN BE USED AS AN ORAL MEDICINE. IN THIS STUDY, THE STRAIN OF PENICILLIUM CHRYSOGENUM (CCRC 31619 OR ATCC 28089) WAS USED TO PRODUCE PENICILLIN V. FIRST OF ALL, THE EFFECT OF INITIAL PH VALUES AND DIFFERENT CARBON SOURCES ON THE PENICILLIN V PRODUCTION WAS EXPLORED. ONCE THE OPTIMUN INITIAL PH VALUE AND CARBON SOURCES HAVE BEEN DETERMINED, THEY WOULD BE USED FOR LATER EXPERIMENT. SECONDLY, THE EXPERIMENT WAS CARRIED OUT IN A BATCH FERMENTOR BY USING MEDIA WITH AND WITHOUT YEAST EXTRACT (YE). THE ROTATIONAL SPEED OF IMPELLER IN THE FERMENTOR WAS ANOTHER FACTOR BEING TAKEN INTO CONSIDERATION. THE SPORE CONCENTRATION WAS 7.3*106 SPORES/ML FOR THE FLASK CULTURE UNDER VARIOUS PH VALUES (6.0, 6.5 AND 7.0). THE CULTURE HAD HIGHEST PENICILLIN V PRODUCTION, REACHING 0.15 G/L UNDER AN INITIAL PH VALUE OF 6.5. THE CONSUMPTION RATES OF GLUCOSE AND (NH4)2SO4 WERE ALSO THE HIGHEST FOR THE CASE OF PH 6.5. THEN, PH 6.5 WAS FIXED WHEN DIFFERENT CARBON SOURCES INCLUDING GLUCOSE, SUCROSE AND LACTOSE, WERE UNDER CONSIDERATION. EXPERIMENTAL RESULTS SHOW THAT GLUCOSE IS BETTER THAN SUCROSE OR LACTOSE AS A CARBON SOURCE FOR PRODUCING BIOMASS AND PENICILLIN V. THEREFORE, PH 6.5 AND GLUCOSE WERE THE BEST COMBINED CONDITION FOR THE FLASK CULTURE. FOR THE BATCH CULTURE IN A FERMENTOR, THE ADDITION OF YE AS A NITROGEN SOURCE COULD LEAD TO HIGHER PRODUCTION OF BIOMASS AND PENICILLIN V. HOWEVER, THE EXISTENCE OF YE ALSO LEADS TO THE DECREASE OF BIOMASS AND PENICILLIN V TOWARD TO THE END OF CULTIVATION. THIS PHENOMENON MIGHT BE DUE TO THE CELLULAR AUTOLYSIS IN THE CULTURE. BECAUSE THE COMPOSITION OF YE IS NOT WELL DEFINED AND MIGHT AFFECT THE EXPERIMENTAL RESULTS IN SOME WAYS, (NH4)2SO4 WAS USED AS THE ONLY NITROGEN SOURCE FOR LATER CULTIVATION OF PENICILLIUM CHRYSOGENUM IN A FERMENTOR. THE D.O. (DISSOLVED OXYGEN) VALUE INCREASED AS THE AGITATION RATE INCREASED. THREE RATES, SAY 150, 200 AND 350 RPM, WERE SELECTED FOR THE EXPERIMENT. THE HIGHEST BIOMASS AND PENICILLIN V CONCENTRATIONS WERE 8.7 G/L AND 0.4 G/L, RESPECTIVELY, FOR THE AGITATION RATE OF 350 RPM. HOWEVER, THE INCREASE OF THE AGITATION RATE WILL INCREASE THE SHEAR STRESS WHICH IS HARMFUL TO THE MICROBIAL GROWTH. IN THIS STUDY, THE INFLUENCE OF SHEAR STRESS SEEMS NOT SIGNIFICANT, THIS MAY BE DUE TO THE RATES OF 350 RPM IS STILL HIGH ENOUGH TO PRODUCE HARMFUL SHEAR STRESS TO MICROORGANISM. PENICILLIUM CHRYSOGENUM IS AN AEROBIC FUNGAL, AND MUST BE CULTIVATED IN AN ENVIRONMENT WITH HIGH DISSOLVED OXYGEN. AERATED AIR CAN ENHANCE THE DISSOLVED OXYGEN IN THE MEDIUM IF CULTIVATED IN A FERMENTOR, AND HENCE THE PENICILLIN V PRODUCTION IS HIGHER THAN THAT OF PENICILLIN V PRODUCED IN A SHAKER FLASK CULTURE. NOTE THAT THE LACK OF DISSOLVED OXYGEN MAY LIMIT MICROBIAL GROWTH AND PENICILLIN V PRODUCTION IF CULTIVATED IN A SHAKER FLASK.

Keywords : Penicillium chrysogenum ; penicillin V ; shaker flask culture ; batch fermentation ; initial pH value ; carbon source ; YE ; agitation rates

Table of Contents

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