Rheology of *Penicillium chrysogenum* Pellet Suspensions

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**Summary.** Shear diagrams of *Penicillium chrysogenum* pellet suspensions were measured in a Couette viscosimeter, developed for pellet suspensions as functions of the pellet volumetric fraction. At low pellet concentrations the suspensions show pseudo-plastic behavior. The variation of the apparent viscosity, \( \eta_s \), with the relative pellet volume fraction \( E_X/E_m \) and the shear rate \( D \) can be described by the relationship:

\[
\eta_s = \left( \frac{E_X/E_M}{1 - E_X/E_M} \right)^2 C^2 D^{-b(E_X/E_m)}
\]

where \( E_X \) is the pellet volume fraction and \( E_m \) the maximum pellet volume fraction in the medium, \( C \) and \( b = 1 \) are constants.

The relationship holds true in the range \( \eta_s \geq 20 \eta_0, D \geq 10 \text{ s}^{-1} \) and \( E_X/E_m < 0.08 \), where \( \eta_0 \) is the viscosity of the cellfree medium.

**Introduction**

Several industrial processes use moulds in submersed cultures. The physical properties of these moulds are dependent on the form of the mycelium: filamentous and pellet form. The filamentous structure is accompanied by high viscosity when the hyphae are long and branched. Pellet suspensions are usually less viscous. König et al. (1982) have shown that penicillin production is possible by using *Penicillium chrysogenum* in air lift tower loop reactors, if the high viscosity of the filamentous mould is reduced by using pellet suspensions.

Van Suijdam (1980) investigated the rheology of *P. chrysogenum* pellet suspensions in a shear rate range below \( D = 20 \text{ s}^{-1} \) and recommended the use of the Casson model:

\[
\eta_s = \frac{\tau}{D} = K_c^2 + \frac{\tau_0}{D} + 2 K_c \left( \frac{\tau_0}{D} \right)^{0.5}
\]

for these suspensions. In Eq. (1) \( \eta_s \) is the apparent viscosity of the suspension, \( \tau \) the shear stress, \( D \) the shear rate, \( \tau_0 \) the yield stress and \( K_c \) the limiting viscosity at a high \( D \). For the influence of the pellet volume fraction \( E_X \) on \( K_c \) and \( \tau_0 \) the following relationships were recommended (Suijdam 1980):

\[
K_c = c_1 E_X + c_2
\]

\[
\tau_0 = c_3 (E_X/d)^3
\]

where \( c_1, c_2 \) and \( c_3 \) are the constants and \( d \) is the mean pellet diameter. Comparable measurements at higher shear rates have not been published yet. Metz et al. (1979) gave an excellent review on the rheology of the mould suspensions, mostly in filamentous form. Early measurements in this field are reported by Deindoerfer et al. (1955, 1960a, 1960b) and detailed investigations concerning the filamentous structure have been done by Metz (1976). Yeast suspensions were investigated by El-Temtamy et al. (1982) in the range of \( 10^4 < D < 10^5 \text{ s}^{-1} \). They recommended Eq. (4).

\[
\eta_s = a e^{bE_X} D^m
\]

where \( a \) and \( b \) are constants. No dependence of the flow behavior index \( m \) on \( E_X \) was found.

Reuss et al. (1979) also investigated yeast suspensions in the range of \( 0 < D < 200 \text{ s}^{-1} \) and found no deviation from the newtonian behavior. They recommended the use of Eq. (5):

\[
\eta_s = \eta_0 = \frac{1}{1 - (hX_s)^a}
\]
In Eq. (5) are $\eta_0$ the viscosity of the supernatant, $\eta_r$ the relative viscosity, $h_\text{p}$ the packing factor, $a$ the constant. Simmons et al. (1976) used the relationship (6):

$$\eta_r = \frac{2.02}{1 - E_X} \eta_0 + 1.36 E_X.$$  

Equation (6) assumes that $\eta_r$ consists of two terms: One of them depends on $\eta_0$ and the other is independent of $\eta_0$.

However, for $E_X = 0$ there is a contradiction in Eq. (6). At high $E_X$ values Eq. (6) does not work either (El-Temtamy et al. 1982).

A large number of relationships for the relative viscosity of smooth hard sphere suspensions was reviewed by Rutgers (1962). He found 98 different relationships in literature. Well-known are the relationships of Einstein (1906, 1911, 1920) and Vand (1948), which do not apply to high solid concentrations. The relationship of Eilers (1941)

$$\eta_r = \left(1 + \frac{2.5 E_X}{2(1 - E_X/E_m)}\right)^2$$  

often proved to be true.

Equation (7) behaves well for $E_X \rightarrow E_m$ (maximum packing density) as well as for $E_X \rightarrow 0$. In the first instance $\eta_r$ becomes infinite, in the second instance $\eta_r$ approaches the Einstein relationship (8)

$$\eta_r = 1 + 2.5 E_X.$$  

A great variety of equipment is used for the viscosity measurements of particle suspensions (Langer and Werner 1981). Rotational viscosimeters are fairly popular, because the sedimentation of large particles above a critical shear rate can be blocked (Padberg 1982). Only the capillary, the falling sphere, the Couette and the torsional pendulum methods furnish true fluidity values (Meskat 1957). For the falling sphere and torsional pendulum viscosimeters the flow equation,

$$\frac{dv}{dy} = f(r),$$  

which is independent of the equipment used, cannot be calculated from the shear diagram. In a capillary viscosimeter the sample volume is extremely large, if the adverse wall effects are to be eliminated. A Couette viscosimeter can only be used, if some important conditions are fulfilled first (Schügerl et al. 1961). Impeller systems can only yield relative viscosity values. This is why the Couette viscosimeter was used for the current investigations.

**Experimental Methods**

Organism and Cultivation. *Penicillium chrysogenum* (S1) donated by Hoechst AG, was used for the investigations. The medium compositions are given in Tables 1 and 2.

The first preculture was inoculated with a spore concentration of $10^7$ m$^{-3}$ and was cultivated in a stirred 500-ml vessel with 180 ml medium volume at 25°C for 72 h. The second preculture was inoculated with the entire first preculture and was cultivated in a 25-l stirred tank (b10 Braun, Melsungen) at 550 rpm and 25°C with 16 l medium volume for 40 h. The main culture was carried out in a 94-l air lift tower reactor with up to 80 l medium volume at 25°C in fed-batch operation. The inoculum for the main culture amounted to $1 \times 10^8$. A mixture of lactose and glucose (2–3 weight ratio) as well as soy oil was fed continuously and precursor (K-phenoxiacetate), urea, ammonium sulfate, and pH-correction were added to the medium from time to time. After a cultivation time of 70 h, 50 g of meat extract per day was fed into the reactor. The pH-value was kept between 5.8 and 6.2. For the viscosity measurements the samples were drawn at two different cultivation times: 26 and 100.6 h.

**Viscosity measurements**

The viscosity measurements were carried out in a Couette viscosimeter constructed for pellet suspensions. The width of the annular gap was 5 mm. The dimensions of the inner and the outer

<table>
<thead>
<tr>
<th>Table 1. Medium composition of the first and second preculture</th>
<th>500 ml vessel (g · l$^{-1}$)</th>
<th>Stirred tank (g · l$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cornstepp (solid)</td>
<td>15.0</td>
<td>15.0</td>
</tr>
<tr>
<td>Lactose monohydrate</td>
<td>40.0</td>
<td>10.0</td>
</tr>
<tr>
<td>Glucose monohydrate</td>
<td>0.8</td>
<td>30.0</td>
</tr>
<tr>
<td>CaCO$_3$</td>
<td>1.0</td>
<td>5.0</td>
</tr>
<tr>
<td>(NH$_4$)$_2$SO$_4$</td>
<td>2.0</td>
<td>2.0</td>
</tr>
<tr>
<td>KH$_2$PO$_4$</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>Soy oil</td>
<td>1.0</td>
<td>—</td>
</tr>
<tr>
<td>Carbopol 934</td>
<td>5.0</td>
<td>—</td>
</tr>
<tr>
<td>Desmophen 3600</td>
<td>0.28 ml · l$^{-1}$</td>
<td>0.28 ml · l$^{-1}$</td>
</tr>
</tbody>
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<thead>
<tr>
<th>Table 2. Medium composition of the main culture (tower reactor)</th>
<th>Tower reactor (g · l$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pharmamedia</td>
<td>7.5</td>
</tr>
<tr>
<td>Lactose monohydrate</td>
<td>2.5</td>
</tr>
<tr>
<td>(NH$_4$)$_2$SO$_4$</td>
<td>10.0</td>
</tr>
<tr>
<td>KH$_2$PO$_4$</td>
<td>5.0</td>
</tr>
<tr>
<td>CaSO$_4$-dihydrate</td>
<td>0.0625</td>
</tr>
<tr>
<td>MgSO$_4$-heptahydrate</td>
<td>0.250</td>
</tr>
</tbody>
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