Sieve Plate Hole Pressure Fluctuations: Bubble Formation in Aerated Reactors

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The introduction of bubbles into a liquid-phase medium is common to a number of critically important mass-transfer operations; however, there may be none where the characteristics of bubble formation are as important as in the operation of aerated bioreactors. This is particularly true in the large-scale culture of mammalian cells for the production of therapeutic agents, where cellular susceptibility to hydrodynamic stress is well-documented. In previous reports Rainbolt and Glasgow (1993), Glasgow and Rainbolt (1994), and Glasgow et al. (1994) have characterized both bubble formation transitions and energetic events occurring immediately after bubble detachment from sieve plate spargers in an air-lift reactor. These efforts have demonstrated that very large forces can arise from interfacial motions in the form of retraction of the trailing surface of elongated bubble structures. It was also shown that this phenomenon can occur repetitively at frequencies exceeding 10 Hz for particular operating conditions. These findings warrant a more detailed look at pressure fluctuations accompanying bubble formation, with a view towards identification of operational parameters that could result in a reduction of stresses acting upon suspended entities.

Bubble formation has been intensively studied because of its practical importance to the process industries. Clift et al. (1978) reviewed work that had been carried out for bubble formation under both constant flow and constant pressure conditions. They noted that bubble formation at orifices is disconcertingly complex, with bubble volume depending upon as many as ten or more parameters. Simplifications are possible in some circumstances. For example, if the volume of the chamber or reservoir immediately upstream from the orifice is large relative to the bubble volume, then the variation in gas flow does not affect chamber pressure. Complete and physically accurate modeling of the bubble formation process has proven to be elusive because: (1) at higher gas-flow rates the flow through the orifice is turbulent; (2) the shape of the forming bubble may not be spherical; (3) the induced flow in the liquid medium is turbulent; and (4) inertial forces in the gas may not be negligible. As a result, early work in this area was generally correlational (see the compilations for both bubble volume and bubble frequency provided by Davidson and Amick (1956)). Other investigators constructed macroscopic force balances treating the bubble as a complete entity; Hughes et al. (1955), for example, included buoyancy, pressure, drag, and surface forces, and set those terms equal to the rate of change of gas (bubble) momentum. According to the static (Laplacian) balance, interior bubble pressure (p_b) and bubble size (r) are related by

$$p_b - p_f = \frac{2\sigma}{r}, \tag{1}$$

where σ is the surface tension. Kupferberg and Jameson (1969) used potential flow theory to develop equations for the motion of a growing spherical bubble. Neglecting variations in interior bubble pressure with respect to angular position, they found

$$p_b - p_f = \rho\left[r\frac{d^2r}{dt^2} + \frac{3}{2}\left(\frac{dr}{dt}\right)^2 - gs\right] + \frac{2\sigma}{r} \tag{2}$$

where s is the vertical distance between the center of the bubble and the orifice.

Murhammer and Goochee (1990) reported that *Spodoptera frugiperda* (insect) cells appeared to be damaged by events occurring in the vicinity of gas distributors in aerated bioreactors. They described three possible sources of disruptive energy: (1) rapid interfacial motion at the trailing (bottom) bubble surface accompanying and following detachment; (2) bubble oscillation following closure (detachment); and (3) interfacial velocity resulting from the pressure drop across the gas distributor. They proposed the concept of a "killing zone" in the vicinity of the sparger in aerated bioreactors, where interactions between cells and small eddies might result in loss of viability. Since the events that accompany bubble formation in aerated reactors are inherently periodic, it is important to note that pulsatile conditions have been shown to lead to physiologic changes in cell structure and function. Frangos et al. (1985) found that pulsatile conditions led to greater production of prostacyclin by human endothelial cells. Davies et al. (1986) and Levesque et al. (1989) reported the effects of turbulent and pulsatile stresses, respectively, upon vascular endothelial cells; they described resulting changes in cell loss rate, cell stiffness, and cell proliferation.

We have been concerned with hydrodynamic forces of interfacial origin that might act upon fluid-borne cells in large-scale culture processes. In this regard, Glasgow and Rainbolt (1994) examined interfacial velocities occurring with bubble formation at sieve plate spargers using high-speed macro-videography. This work demonstrated that energetic fluid motions could result from bubble formation and detachment phenomena, especially at intermediate viscosities. Consequently, proper examination of the bubble formation process might lead to improved design and operation of aerated bioreactors, resulting in enhanced culture productivity.

Experimental Procedure

The work described here was carried out with a 3-L acrylic plastic, split-column airlift fermenter. This device has been described in detail by Rainbolt and Glasgow (1993). Air was sparged into the liquid medium through a sieve plate (12-mm thick) with a single, 1-mm diameter hole. A pressure tap was installed by drilling a 3-mm hole through the side of the sieve plate; this tap intersected the sieve plate hole in the center of the plate and at right angles. The (gas) volume of the chamber immediately beneath the sieve plate was 390 cm^3. The liquid media employed for this investigation included distilled water, 24 and 38% sucrose solutions, 84% glycerol solution, and distilled water with ethanol added for surface tension reduction. The gas flow rates employed ranged from about 1 to 75 cm^3/s. For flow rates less than about 7 or 8 cm^3/s, bubble formation was generally intermittent, in which a series of three to six bubbles would be followed by a period of inactivity. This bubbling mode was described by Hayes et al. (1959), among others.

Hole pressure measurements were made with a piezoresistive bridge-type transducer driven at 21 VDC. The output from the transducer was recorded with a Nicolet 4094 digital storage oscilloscope and then transferred off-line to a PC for further analysis. Data of interest included magnitude of the pressure fluctuations accompanying bubble formation, the frequency and energy distribution of those fluctuations, and the maximal rate of change of pressure (or voltage) dP/dt.

Results

Figure 1 illustrates a typical sequence of hole pressure data for distilled water, using gas flow rates (Q's) ranging from 4.5 to 34 cm^3/s. At $Q = 4.5$ cm^3/s, bubble formation is intermittent with perhaps 12 to 15 bubbles generated per second. Note, however, that the largest pressure fluctuation corresponds with the formation of the first bubble in each sequence, and that pressure change has a magnitude of about 3,900 dyne/cm^2. When the flow rate is increased to 9 cm^3/s, bubble formation is continuous at a rate of about 31 Hz, corresponding to a mean bubble volume of about 0.29 cm^3 per bubble or an average diameter of roughly 8 mm. Note also that at $Q = 9$ cm^3/s, the magnitude of the maximum pressure change is approximately 2,300 dyne/cm^2. As the gas-flow rate is further increased, the size of the maximal pressure fluctuation grows monotonically.

In order to assess the effects of liquid-phase viscosity (but comparable surface tensions) upon this behavior, the experi-

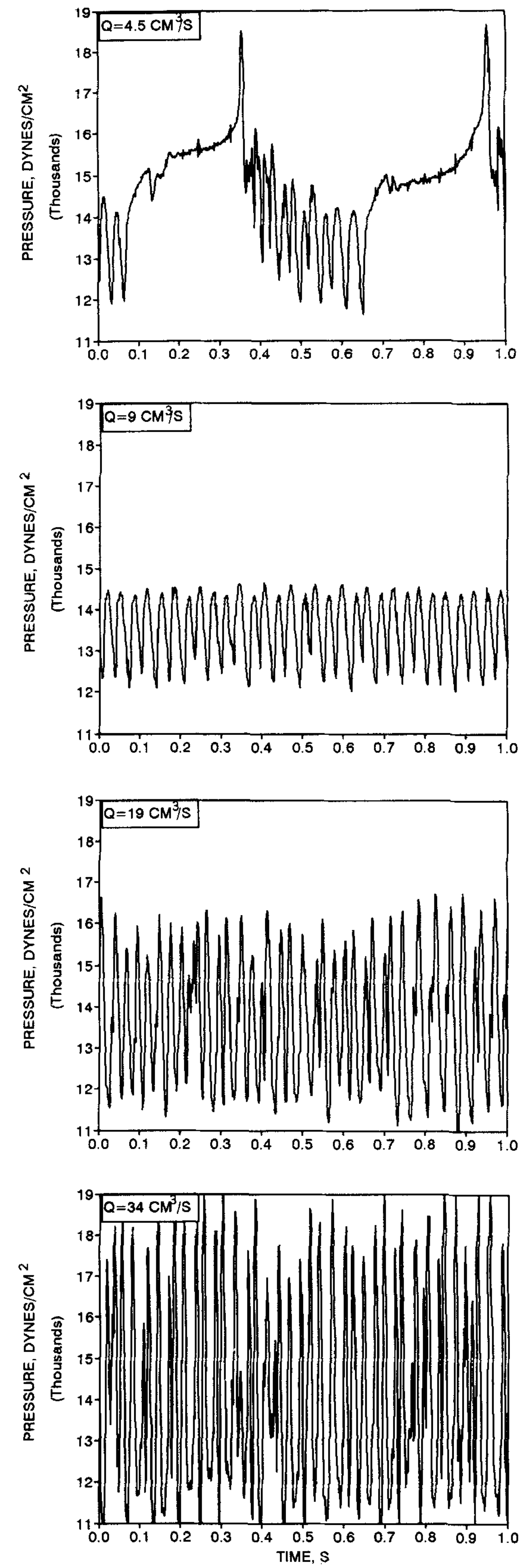

Figure 1. Characteristic hole pressure fluctuations for bubble formation in distilled water.

Bubbles were formed at a 1-mm dia. hole at flow rates ranging from 4.5 to 34 cm^3/s.

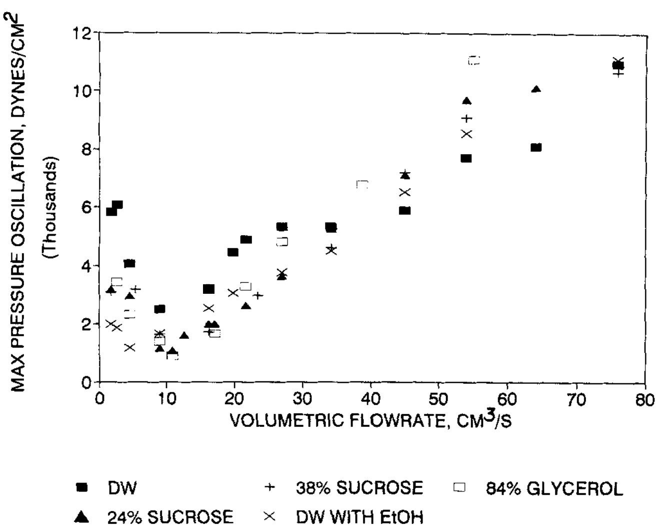

Figure 2. Magnitude of the largest pressure changes accompanying bubble formation for five liquid media.

The differences found at very low gas rates would be significant in the culture of mammalian cells.

ments were repeated using both sucrose and glycerol solutions. Thus the viscosities employed—including distilled water—were in the range of 1 to about 80 cp (80 mPa·s). In addition, an aqueous solution of 53% ethanol was employed to provide some data with significantly lower surface tension (about 28 dyne/cm). The results of this comparison are provided in Figure 2, which shows the magnitude of the maximum observed pressure change (fluctuation) as a function of volumetric flow rate. In all cases, a distinct minimum occurs at the transition from intermittent to continuous bubble formation. Note that at very low flow rates, the maximum observed pressure oscillation for distilled water was on the order of 6,000 dyne/cm^2, while the corresponding value for the ethanol solution was only 2,000 dyne/cm^2. All of the minima are located in the range of flow rates from about 5 to 15 cm^3/s. At intermediate flow rates, perhaps 25 to 50 cm^3/s, there is little difference in hole pressure fluctuation magnitude among the solutions examined. This does not mean, however, that the signal dynamics are comparable; the 24% sucrose solution, for example, produced a hole pressure signal in this range of flow rates that was quite different from that obtained with distilled water. This is illustrated by the power spectra presented in Figure 3. These spectra were obtained by Fourier transformation of time-series (hole pressure) data at fixed gas-flow rate ($Q = 17$ cm^3/s) for three liquid media: distilled water; a 24% sucrose solution; and an 84% glycerol solution. Accordingly, the viscosities and surface tensions were about 1, 2.8 and 72 cp, and 73, 74 and 66 dyne/cm, respectively. Note that virtually all of the signal energy in the case of distilled water is contained between 18 and 24 Hz. For the 24% sucrose solution, major signal energies are located at about 20 and 41 Hz. In contrast, important frequencies for 84% glycerol were 13, 27 and 41 Hz. There is a clear and important implication of these data: If oscillatory disturbances can lead to or produce cell damage, then much greater attention must be paid to the design and operation of gas distributors.

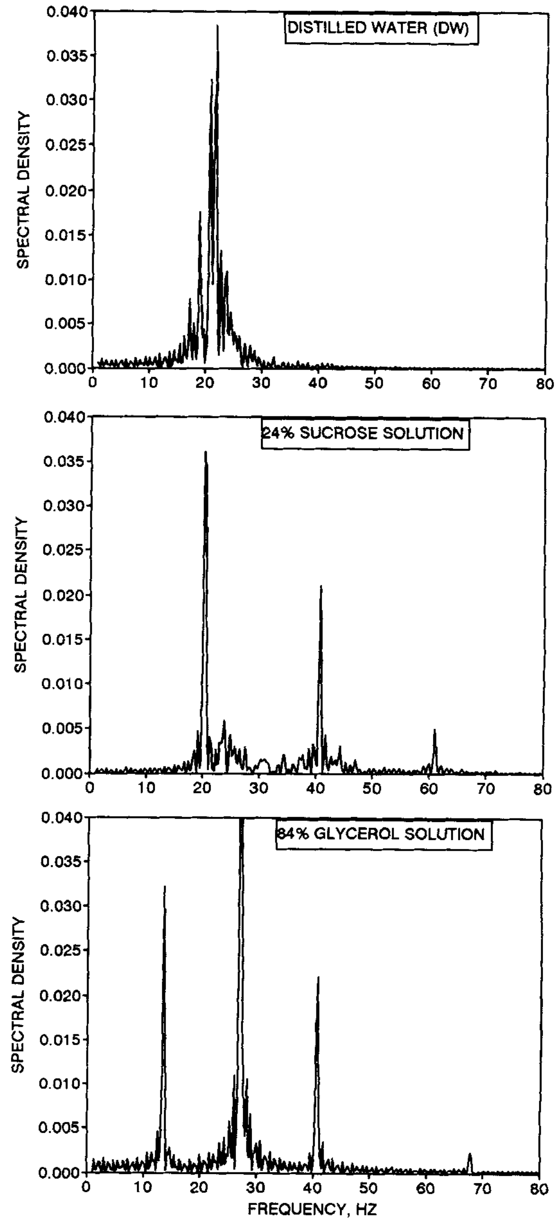

Figure 3. Comparison of power spectra obtained from hole pressure signals at fixed volumetric gas-flow rate $Q = 17$ cm^3/s.

This sequence shows the effect of increasing viscosity at nearly constant surface tension.

Figure 4 shows that in terms of signal energy distribution, a decrease in surface tension brings about a situation that is much more complicated. The addition of ethanol to distilled water results in power spectra in which energy is distributed over a much broader range of frequencies. For example, in the center panel of Figure 4 (with gas-flow rate, $Q = 17$ cm^3/s), there is significant signal energy over the frequency rate 20 to 50 Hz. Furthermore, this situation appears to persist over a broad range of gas-flow rates. These data are at sharp variance with those obtained with distilled water at $Q = 17$ cm^3/s, as depicted in Figure 3.

In addition to pressure signal periodicity, the speed of interface displacement is of major interest; as noted by

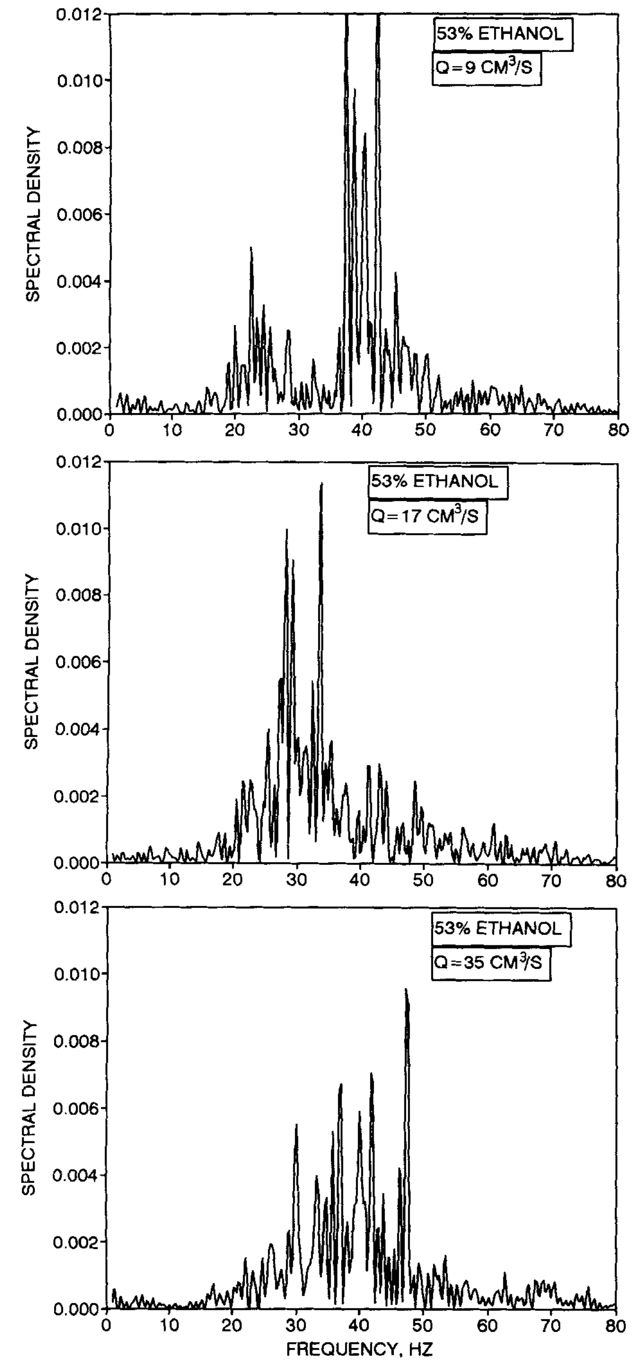

Figure 4. Power spectra obtained with an aqueous solution of 53% ethanol illustrating the effects of a dramatic reduction in surface tension upon pressure signal (energy) distribution.

Murhammer and Goochee (1990), such motions—if sufficiently violent—may have lethal effects upon fluid-borne cells. Since $p_b(t)$ was measured in the work described here, it is possible to solve Eq. 2 numerically to obtain both $r(t)$ and dr/dt, the interfacial velocity. This process was explored during the course of this work for modest gas rates with aqueous sucrose solutions. It was found that the maximum value of dr/dt for 24% sucrose solution with $Q = 17$ cm^3/s was about 29 cm/s. However, Eq. 2 assumes a spherical bubble form that is uncommon in air sparging with sieve plate distributors. For example, a sequence of high-speed video images obtained during bubble formation in an aqueous sucrose solution is provided as Figure 5 with the first of the series, frame 103, appearing in the upper lefthand corner. The bubble surface entering the bottom of frame 103 is forming at the sieve plate hole. Note the deformation of the upper bubble surface in frames 105 through 108. The pointed projection on the upper bubble surface that is apparent in these images moves (momentarily) with a velocity exceeding 80 cm/s. Our previous work concerned with bubble formation at sieve plate spargers makes it clear that bubble shape is significantly affected by proximate, preceding bubbles.

Because of the variability seen in forming bubble shapes, a means of assessing interfacial velocity (during bubble formation) other than numerical solution of force balances is highly desirable. Although dp_b/dt is not directly proportional to dr/dt, it seems that the derivative of the recorded hole pressure signal might be used to provide a semiquantitative comparison of interfacial velocity. The required estimates can be obtained in the following fashion: For cases in which the pressure signal is not intermittent, i.e., bubble formation is not interrupted by periods of inactivity, the maximum pressure fluctuation magnitude is multiplied by the characteristic

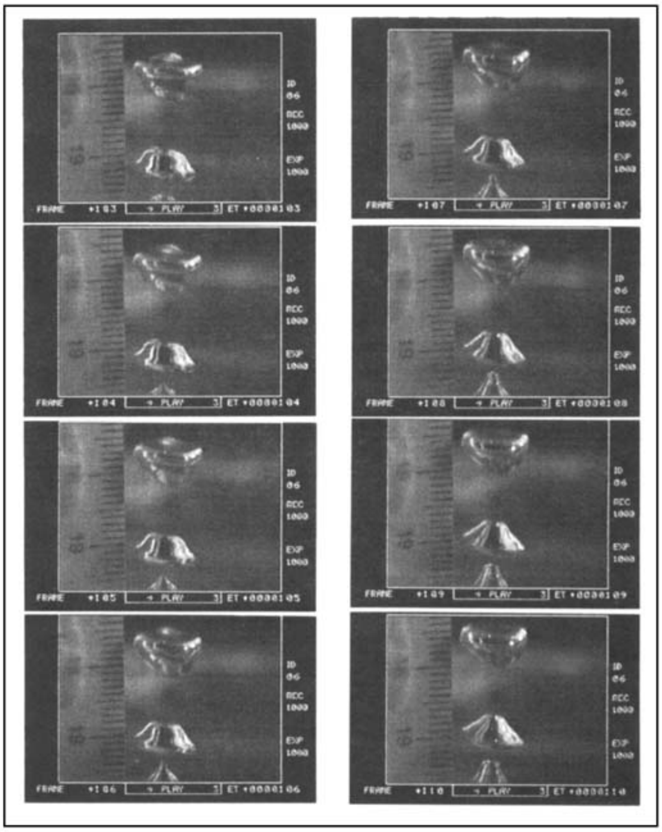

Figure 5. Sequence of high-speed video stills illustrating the deformation of the upper surface of an air bubble forming in an aqueous sucrose solution.

At intermediate gas rates, such processes result in interfacial velocities that greatly exceed values of dr/dt predicted by force balance models for spherical bubbles.

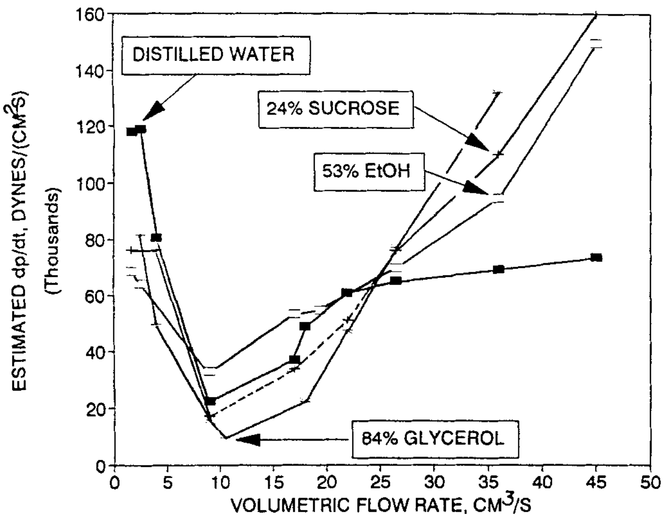

Figure 6. Estimated values of $(dp/dt)_{max}$ for four different liquid media over a wide range of gas-flow rates.

These data serve as semiquantitative indicators of interfacial velocity.

frequency and divided by 2. This simple procedure provides a means for quick, approximate comparison of energetic interfacial motions associated with bubble formation at sieve plate spargers. The data presented in Figure 6 show that, compared to distilled water, increased viscosity results in a decrease in dp_b/dt at intermediate gas-flow rates. A substantial decrease in surface tension (obtained with the addition of ethanol) yields larger values of this derivative at intermediate gas rates. The minimum observed values of dp_b/dt for bubble formation at a 1-mm hole in a plate sparger, occurred at gas flow rates between 9 and 17 cm^3/s.

Conclusions

This work has shown that energetic interfacial motions accompanying bubble formation at sieve plate spargers can be partly controlled through manipulation of flow rate. In general, the gas-flow rate that marks the transition between intermittent and continuous bubbling probably transmits the smallest stresses to the liquid phase. While the stresses at very low gas rates are relatively large, it should be remembered that they occur intermittently (effectively at very low frequencies). However, it is also apparent that the characteristic frequencies associated with bubble formation are quite narrow with distilled water at intermediate gas rates, while significant decreases in surface tension result in a much broader distribution of signal energy or power. Increased medium viscosity (at intermediate gas-flow rates) results in sharply defined peaks in (hole pressure) signal energy at typically two or three frequencies. The results of this work make it clear that decreased surface tension is not a panacea for cell damage that might result from bubble formation processes. In fact, decreased surface tension will make it more difficult to design and operate aerated reactors for culture of cells susceptible to pulsatile disturbances; in addition, decreased surface tension will actually increase the velocity of interfacial motions at intermediate gas rates. The only conditions for which dramatic reductions in surface tension appear to be beneficial (with respect to transmission of stresses to the liquid phase) would be at extremely low gas rates for which bubble formation occurs intermittently (a sequence of bubbles followed by a period of inactivity).

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