Hazards of Mouth Pipetting*

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Introduction

Some form of pipette has probably been used as a basic tool by scientists as long as there have been laboratories. The word "pipette" was apparently introduced into the French language in the 1830's. From 1860 on, the pioneers of bacteriology frequently referred to the use of pipettes. In the 1870's it became common practice to plug the oral end of pipettes with cotton wool. Although the use of pipettes in the early chemistry laboratories undoubtedly led to accidental aspiration of undesirable toxic and poisonous substances, the first recorded laboratory infection due to mouth pipetting occurred in 1893. Kisskalt[2] reported the case of a physician who accidentally

Pike[6] reported that 34 of 1342 laboratory infections occurring between 1930 and 1950 were due to mouth pipetting. Ten of 641 infections gathered from the world literature by Pike *et al*[4] for the years 1951 to 1963 were due to accidental aspiration while pipetting.

In addition to infections, it is obvious that chemical burns, poisonings, and other types of injuries may be caused by accidental aspiration through pipettes. Table I shows the reported accidents due to mouth pipetting at two large research institutions during three-year periods.

TABLE I. Mouth Pipetting Accidents at Two Research Institutions

Aspirated Material	Number of Accidents Institute A 1954-1956	Institute B 1959-1961

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Subsequent surveys of laboratory infections following the turn of the century produced ample evidence that mouth pipetting was a frequent cause of accidental infection among laboratory workers. In 1915 Paneth[3] reviewed 57 laboratory accidents that had resulted in 47 infections. More than 40 percent of the infections were attributed to mouth pipetting.

Accident Statistics

Early identification of the hazard of mouth pipetting undoubtedly prompted many laboratory workers to use bulbs, tubes, or other pipettor devices. It is therefore rather surprising that today, 50 years later, mouth pipetting of infectious or toxic fluids is still accepted practice in many laboratories. Approximately 17 per cent of 921 infections reported in the world literature between 1893 and 1950 were due either to oral aspiration through pipettes or to splashes of culture fluids into the mouth. In the U. S., Sulkin and Pike[6] reported that 34 of 1342 laboratory infections occurring between 1930 and 1950 were due to mouth pipetting. Ten of 641 infections gathered from the world literature by Pike *et al*[4] for the years 1951 to 1963 were due to accidental aspiration while pipetting.

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Aspirated Material	Number of Accidents Institute A 1954-1956	Institute B 1959-1961
Infectious cultures or suspensions	8	12
Acids and alkalies	17	1
Toxic solvents	2	
Poisons	1	
Radioactive materials	1	
	29	13

The seriousness of the mouth pipetting accident with infectious cultures is illustrated by the relative frequency with which it results in infection. According to Paneth's 1915 publication,[3] one infection occurred for each three known pipetting accidents. During the period 1958-1962 both of two mouth pipetting accidents reported at the U. S. Army Biological Laboratories resulted in infection. Over a longer time the records show that the ratio of infections to reported accidents resulting from oral pipetting was 1:5. This can be compared with a ratio of 1:21 for the frequency with which infections resulted from all known accidents.

Specific Hazards of Mouth Pipetting

The two major hazards in mouth pipetting were explained by Paneth[3] and later by Wedum[7]:

1. Mouth pipetting frequently results

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in accidental aspiration of the fluid in the pipette. Pipettes plugged with cotton do not consistently prevent this hazard because overzealous sucking pulls the plug into the mouth along with the fluid in the pipette.

2. Even when aspiration does not occur, contamination of the mouthpiece from one's own contaminated finger can result in oral contamination.

In addition to these hazards, Bloom[1] recently has shown that with radioactive solutions there is a danger of inspiration of vapors through unplugged pipettes. Using a syringe to simulate mouth action, Bloom showed that significant amounts of tritium oxide were detectable in the air aspirated from unplugged pipettes. Each pipette aspiration carried from 5 to 70 millimicroliters of the solution in the pipette to a hypodermic syringe above the pipette. Constant pipetting obviously can result in significant transfer of vapors from the solutions being used.

Experiments with Unplugged Pipettes

We have repeated Bloom's procedure using broth cultures of *Serratia marcescens* (1×10^9 cells per ml) and *Bacillus subtilis* var. *niger* (3×10^{10} cells per ml). In each test, a 10-ml syringe mounted vertically on a stand was used to simulate mouth pipetting of 10 ml of culture into a 10-ml pipette. After 10 mixing cycles, the syringe was removed, rinsed with sterile physiological saline and the saline was added to culture plates to assay for viable organisms. Recovery of organisms from the syringe provided evidence that microbial aerosols had been produced by the mixing procedure and had escaped through the unplugged proximal end normally held in the mouth. A total of 120 trials was conducted with each species of bacteria. Each trial consisted of 10 mixes of the culture with the pipette. The results are shown in Table II.

TABLE II. Aerosols Coming Through the Unplugged Mouthpieces of Pipettes During Tests Simulating Mouth Pipetting

Number of Organisms Recovered in the Syringe	Number of Tests	
	S. marcescens	*B. subtilis*
300 or more	14	7
100-299	0	4
11-99	9	23
1-9	18	35
None	79	51
	120	120

With *S. marcescens*, aerosols were detected in the syringe in 41 of 120 trials (34 per cent). With *B. subtilis*, 69 of the recovery plates (58 per cent) showed the test organisms. Although there was considerable variation in the amounts of aerosol recovered, the average number of organisms detected from the positive tests was 116 for *S. marcescens* and 61 for *B. subtilis*. It is clear from these results that if non-plugged pipettes are used in mouth pipetting there is a possibility of gradual oral contamination even in the absence of accidental aspiration of fluid.

Avoiding the Hazards

Compared with the equipment and procedures required to avoid other types of microbiological laboratory hazards, the method of avoiding pipetting hazards is so elementary, so simple, and so well-recognized that it seems redundant to mention it. However, continued accidents and infections in laboratories illustrate, even today, that there is a lack of acceptance of the simple precautionary measures needed.

In 1915 Paneth[3] concluded that use of a rubber bulb as a pipettor device would avoid the major hazards of infection and that the use of a rubber hose attached to a pipette would avoid direct oral aspiration of fluids but may not prevent oral contamination from finger contamination. In 1950 Schafer[3] stated: "The chief source

of laboratory infections during the bacteriological-serological diagnosis of typhoid fever is due to the pipetting of live cultures." In the same year Wedum[7] described a number of devices for pipetting in the microbiological laboratory.

Several large laboratory institutions, among which are the U. S. Army Biological Laboratories and the U. S. Naval Biological Laboratories, have instituted regulations forbidding mouth pipetting of infectious or toxic fluids. In several European countries federal regulations applying to all medical laboratory workers also prohibit mouth pipetting of dangerous substances. In one country infection due to mouth pipetting is grounds for denial of work-loss compensation. Regardless of regulations, however, adequate avoidance of the pipetting hazards is achieved only when there is understanding and acceptance of the necessary precautions by every laboratory worker who handles dangerous substances.

The rules to follow are simple:

1. DO NOT MOUTH PIPETTE INFECTIOUS OR TOXIC FLUIDS.
2. USE A PIPETTOR DEVICE FOR PIPETTING.

Conclusions

Mouth pipetting of infectious or toxic fluids presents three hazards.

1. Accidental aspiration of the fluid in the pipette. This occurs even when pipettes are plugged with cotton.

2. Aspiration of vapors or of aerosols from the fluid when mouth pipetting with unplugged pipettes.

3. Oral contamination following the placing of a contaminated finger on the proximal end of the pipette.

Accidents due to mouth pipetting have been recognized as a source of laboratory infection for at least 50 years. A significant number of infections and injuries are still caused by mouth pipetting.

The hazards of mouth pipetting are avoided by the use of an adequate pipettor device.

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