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TAPE APPLICATION: A JAR SEALING METHOD FOR REDUCING ETHANOL EVAPORATION IN FLUID-PRESERVED COLLECTIONS

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Abstract.—Evaporative loss is a major concern for fluid-preserved collections. Evaporation leads to a decrease in preservative concentration and eventually to specimen desiccation. Evaporative loss from small jars (<1 liter capacity) was observed in the Fish Collection at the Canadian Museum of Nature. Research into reducing rates of evaporative loss was initiated since small jars account for about 40% of the 100,000 jars in the Fish Collection. Five different tapes were tested for their sealing properties at the jar-lid junction of 375 ml jars. Teflon® tape was applied to the jar necks in 3 different ways. Adhesive tapes (aluminum foil, polyethylene-rubber, polyethylene-acrylic and polypropylene-acrylic) were applied over the outside of the jar-lid junction. Jars were initially filled with 10 ml of a solution of 70% v/v ethanol/water. Evaporative loss was assessed by calculating the weight reduction of the jars. Regression graphs showing weight loss over time and a grouped box plot were used to compare the different test groups. Results indicated that the rate of evaporative loss was reduced significantly with the application of polypropylene/acrylic-adhesive tape over the jar-lid junction. The acrylic adhesive was tested for stability in the presence of ethanol solutions at room temperature (20°C) and at 50°C.

INTRODUCTION

Evaporative loss of preservatives such as ethanol-water and isopropanol-water solutions is a serious concern in the curation of any fluid-preserved collections. The Fish Collection at the Canadian Museum of Nature holds over 100,000 jars. About 40% of these are small jars (<1 liter capacity). Difficulty in obtaining high quality containers with a tight seal has led to loss of large amounts of preservatives throughout the years. Recognition of this problem resulted in a labor intensive collection maintenance program.

Prior to 1986, most specimens were kept in glass jars with metal or Bakelite lids. The preservative was a nominal 50% volume/volume (% v/v) isopropanol/water solution. A solution of 50% v/v isopropanol/water was used for topping up jars when the fluid levels were low. A gradual decrease in the solution concentration resulted. In 1986, after the acquisition of a PAAR density meter (Appendix), the concentration of the isopropanol/water solutions was checked. It was revealed that the majority of specimens in the Fish Collection were preserved in isopropanol solutions ranging from 6% v/v to 40% v/v, with a mean of about 20% v/v. At that time, most metal and Bakelite lids were replaced by white, polypropylene lids. Bakelite lids contribute to high rates of fluid-preservative loss since they tend to loosen with temperature fluctuations and to become brittle and crack (Fink *et al.,* 1979; Simmons, 1990; Suzumoto, 1992). The preservative was also changed through a graded series to a 70% v/v ethanol/water solution. Ethanol is less toxic than isopropanol (Commercial Alcohols Inc., 1994; Canadian Center for Occupational Health and Safety, 1993) and may be better suited for use in natural history collections (de Moor, 1990; Emerson and Ross, 1965). Nevertheless, fluid-preservative loss was still evident. Through consultation with other institutions such as the Huntsman Marine Laboratory (New Brunswick), we

learned that sheet-polyethylene lid liners were widely used to obtain a better seal. Polyethylene foam liners, glued inside a polypropylene lid, were not available in Canada at that time. Since then, Njie and Waller (1993) have shown that polyethylene foam liners reduce evaporation by about three folds when compared to sheet-polyethylene liners. In 1987, sheet-polyethylene liners 0.5 mm thick were added to the polypropylene lids. Five years later, more than 25% of the preservative had evaporated from many of the small jars (mainly 250 ml or smaller). A better method for reducing evaporation had to be found.

In 1975, 26 small jars were selected from the collection to test the ability of polyethylene/rubber-adhesive tape to seal the jar-lid junction. After 11 years, the tape was still wrapped tightly around the jars and the polyethylene backing remained flexible; however, the rubber-based adhesive had lost its adhesive property and crumbled upon removal of the tape. Preservative loss was, nevertheless, very low, and the initial concentration of alcohol had not decreased substantially. This observation prompted further investigation in the use of tapes as a practical method of reducing evaporation in fluid-preserved collections. After consultation with technical staff at 3M, samples of the tape previously used, and others with long lasting properties, were obtained for testing. We decided to broaden the experiment by adding other known means of sealing methods such as Teflon® tape and aluminum foil tape.

Materials and Methods

Five different tapes were selected for testing: polytetra-fluoroethene (Teflon®) tape (T), aluminum foil tape (M), polyethylene/rubber-adhesive tape (PER), polyethylene/acrylic-adhesive tape (PEA), and polypropylene/acrylic-adhesive tape (PPA). Acrylic adhesives are of the same formulation for groups PPA and PEA. Also, the polyethylene backing is the same in groups PER and PEA. Teflon® tape was applied three different ways to determine the most efficient application method. Universal Screw Type jars were used (See Appendix for Supplier). The 375 ml size was chosen because it represents the smallest jar size currently used in the Fish Collection. Nine groups of jars were tested. Each group contained 12 jars. One or two jars were selected from each of nine cartons of 12 new jars. The cartons of new jars were listed by the manufacturer as from the same lot. Table 1 describes the tapes, their application, and the test groups.

Each jar was inscribed with its group and jar number using a black wax crayon on the top of its lid, then weighed on a Mettler AT400 balance (Appendix) before adding the 70% v/v ethanol/water solution. The balance's sensitivity is 0.1 mg over the 400 g range. Group B jars were weighed with jar and lid, groups L, PEA, PER, PPA and M with jar, lid, and liner, and groups T, TT and TO with jar, lid, liner, and Teflon® tape. Using a pipette, ten millilitres of 70% v/v ethanol/water were added to each test jar. Lids (manually placed on jars) were tightened with a torque-limiting device designed and constructed by the museum's conservation staff (Njie and Waller, 1993). The torque applied was approximately 30 Nm which is comparable to that normally obtained by hand. All filled jars were weighed again. Adhesive tapes then were applied to jars in groups PEA, PER, PPA and M. These groups were weighed a third time in order to determine the initial weight of the alcohol. All test jars were stored under a loosely fitting Tyvek® cover (to reduce dust accumulation) on a stainless steel cart.

All jars were weighed weekly. A random number table was used to select the weighing order. Date (day, month) and weighing time (hours, minutes) per group, relative humidity and temperature (from hygrothermograph), and barometric pressure and room temperature were recorded also. Partial pressure of water vapor in the air was calculated from measured relative humidity and temperature values using the table in Weast (1989, p. D-190, Vapor Pressure of Water Below 100°C). Barometric readings were recorded at the start of weighing each group of 12 test jars. The relative humidity and temperature were recorded twice during the weighing: at the beginning and after the fourth group of 12 test jars. The Mettler balance was re-calibrated after every fourth test jar weighed. The weighing experiment

Table 1. Description of tapes, application methods, and test groups.

Test groups	Description
B	Jars with lid* only (Control Group).
L	Jars with lid and liner**.
T	Jars with lid; liner; and 1 layer of Teflon® tape***, wrapped around the thread of the jar neck.
TT	Jars with lid; liner; and 2 layers of Teflon® tape, wrapped around the thread of the jar neck.
TO	Jars with lid; liner; and 2 layers of Teflon® tape, 1st layer wrapped around the thread of the jar neck and the 2nd layer partially overlapping the first and extending over the rim inside the lid.
PER	Jars with lid; liner; and polyethylene/rubber-adhesive tape (3M Product #483), (Appendix), black, 2.5 cm wide, 0.13 mm thick, wrapped 1 1/2**** times around the closed jar at the junction of lid and jar neck.
PEA	Jars with lid; liner; and polyethylene/acrylic-adhesive tape (3M Product #480), (Appendix), 2.5 cm wide, 0.13 mm thick, wrapped 1 1/2 times around the closed jar at the junction of lid and jar neck.
PPA	Jars with lid; liner; and polypropylene/acrylic-adhesive transparent tape (3M Product #5086), (Appendix), 2.5 cm wide, 0.07 mm thick, wrapped 1 1/2 times around the closed jar at the junction of lid and jar neck.
M	Jars with lid; liner; and an aluminum foil adhesive tape, 3.5 cm wide, wrapped 1 1/2 times around the closed jar at the junction of lid and jar neck.

* Lids used for all groups are white polypropylene.
** Liners used for all groups, except B, are 0.5 mm thick polyethylene.
*** Teflon® tape used for groups T, TT, and TO is 13 mm wide and 0.01 mm thick.
**** Tape was wrapped once around the closed jar and extended further, about half the circumference, to strengthen the seal by overlapping the tape.

ran for 16 consecutive weeks. All recorded measurements were entered into a computer using QUATTRO® PRO 4.0 software.

The measured weights of the low density jars were subject to significant variations with changes in air density which, in turn, vary due to changes in barometric pressure, temperature, and relative humidity. This problem was circumvented by converting all measured weights to mass in vacuo using the measured temperature, pressure, and relative humidity (Dean, 1979).

Rates of evaporative loss for each jar type were determined by linear regression. A grouped box plot was used to illustrate the variability between and within test groups.

Jar B7 in group B was broken at the beginning of the experiment. Consequently, all calculations for group B were based on eleven jars.

Results and Discussion

Comparison of results for each sealing method.—Each group was ranked from low to high based on the mean rate of evaporative loss (Table 2). Group TT ranked first (lowest) with a mean rate of evaporative loss of 588 mg/year, closely followed by PPA (649 mg/year), and PER (673 mg/year). Groups M (1,028 mg/year), PEA (1,286 mg/year) and L (1,309 mg/year) showed higher evaporative losses. Finally, groups T (2,018 mg/year), B (3,908 mg/year) and TO (4,527 mg/year) showed the highest rates of evaporative loss. Regression graphs showing the evaporative loss over time were plotted for each group. Figure 1 shows that the best fitted curve for each jar does not clearly intersect zero. Consequently, the precision of the intercept, with 95% confidence interval, was calculated for

Table 2. Mean evaporative loss and 95% confidence interval (CI) for test groups.

Test groups	Evaporative loss and 95% confidence interval	Minimum (mg/year)	Maximum (mg/year)	Mean (mg/year)	Rank
B	Rate of loss	501	11,890	3,908	8
	CI			2,711	
L	Rate of loss	167	6,272	1,309	6
	CI			1,289	
T	Rate of loss	59	15,769	2,018	7
	CI			2,780	
TT	Rate of loss	106	1,203	588	1
	CI			259	
TO	Rate of loss	2,211	7,855	4,527	9
	CI			1,117	
PER	Rate of loss	173	1,885	673	3
	CI			325	
PEA	Rate of loss	285	4,136	1,286	5
	CI			850	
PPA	Rate of loss	332	1,261	649	2
	CI			190	
M	Rate of loss	76	5,378	1,028	4
	CI			894	

each jar (Elliott, 1971). The results demonstrate a zero intercept for most groups. Group TO was the only group showing a non-zero intercept.

A grouped box plot shows the extent of evaporative loss (mg/year) that might occur for each group including their mean and outside values (Fig. 2). The best performing group can be assessed by such factors as the small range in the rates of loss as well as the lowest mean rate of loss. Some of the wide ranges in evaporation rates might be caused by different agents. For example, in Group B, the quality of the glass mold might determine whether a good seal between mouth of jar and lid is achieved. Glass mold quality becomes less of a factor with the addition of a polyethylene liner as seen in Group L, where the mean rate of evaporative loss was reduced by 67%.

Group TO showed a mean rate of evaporative loss greater than that for Group B. In group TO, Teflon® tape overlapped the jar rim. The lid seemed to tear and/or disturb the tape during the tightening process, creating an uneven seal at the jar-lid junction. This would explain the higher mean rate of loss as well as the non-zero intercept seen in the linear regression for this group. The far outside value seen in Group T corresponds to jar T10. It suggests a problem in the application of the tape for Group T. A poor seal can occur when the end of the Teflon® tape extends over the rim of the jar, as was the case with jar T10. The use of Teflon® tape was, therefore, deemed unreliable. Problems with Teflon® tape application cannot be visually detected when lids are in place and can cause high rates of loss. Even though Group TT had the lowest mean rate of evaporative loss and one of the smallest ranges in rate of evaporative loss, we do not recommend its use because of the problem with the other Teflon® tape test groups.

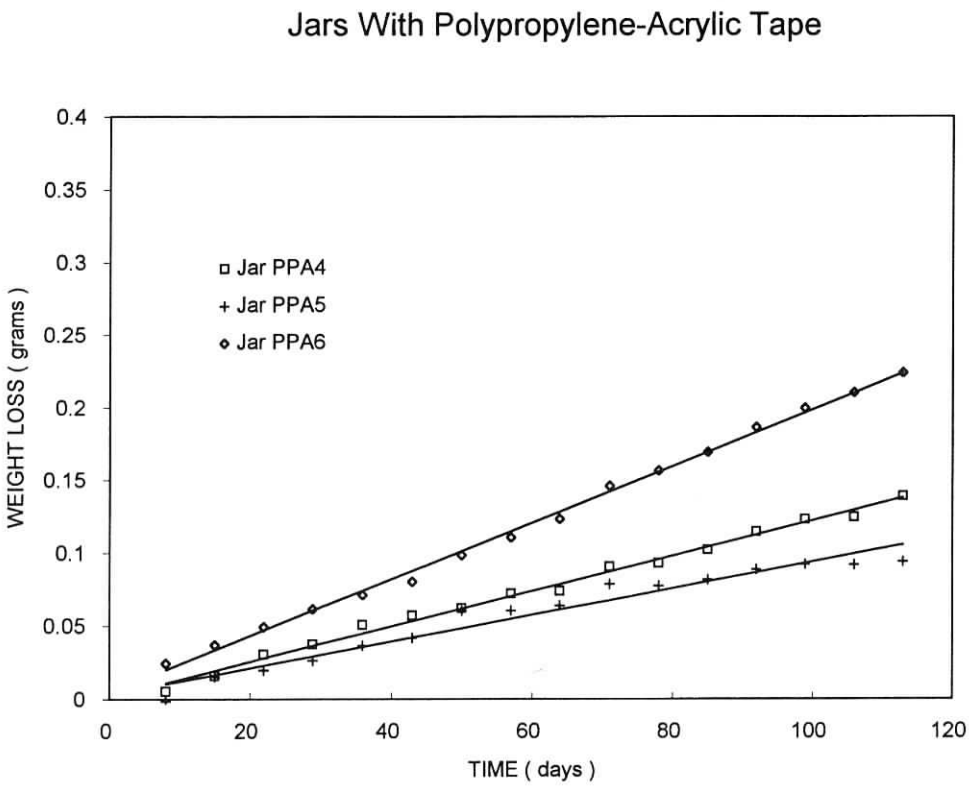

Figure 1. Regression graph showing weight loss (alcohol evaporation) over time for three of the jars (PPA4, PPA5, PPA6) tested with the application of polypropylene/acrylic-adhesive tape.

The aluminum foil tape, group M, did not conform to the jar-lid junction as well as PPA, PER, and PEA tapes. Wrinkles in the foil tape on the jars might have contributed to a greater range in rate of loss. Aluminum tape was also deemed unreliable.

Groups PPA and PER showed good results similar to those obtained for Group TT. Since TT cannot be recommended, the second ranked group, PPA, becomes the first choice. Even though PER showed similar mean rates of loss to PPA, PER is not recommended for long term use because of the deterioration potential of the rubber adhesive. This potential was illustrated by the 1975 experiment with PER tape, in which the adhesive had degraded severely within 11 years. According to Horie (1987) acrylic adhesive is expected to perform much better than rubber adhesive. It resists ageing, an important factor for museums.

Characteristics of polypropylene and polyethylene tapes.—Tests performed by the manufacturer have shown that the acrylic adhesive is strongly resistant to degradation by humidity, heat, and light conditions (3M, 1989a, c). These characteristics have also been confirmed by staff at the Canadian Conservation Institute (pers. comm., J. Tétreault, 1993). In addition, a study on the evaluation of adhesives demonstrated that acrylic adhesives released minimal levels (seen in one case only) or no acid emission at all (Down *et al.,* 1992). The acrylic adhesive, found on PPA, is compatible with museum needs.

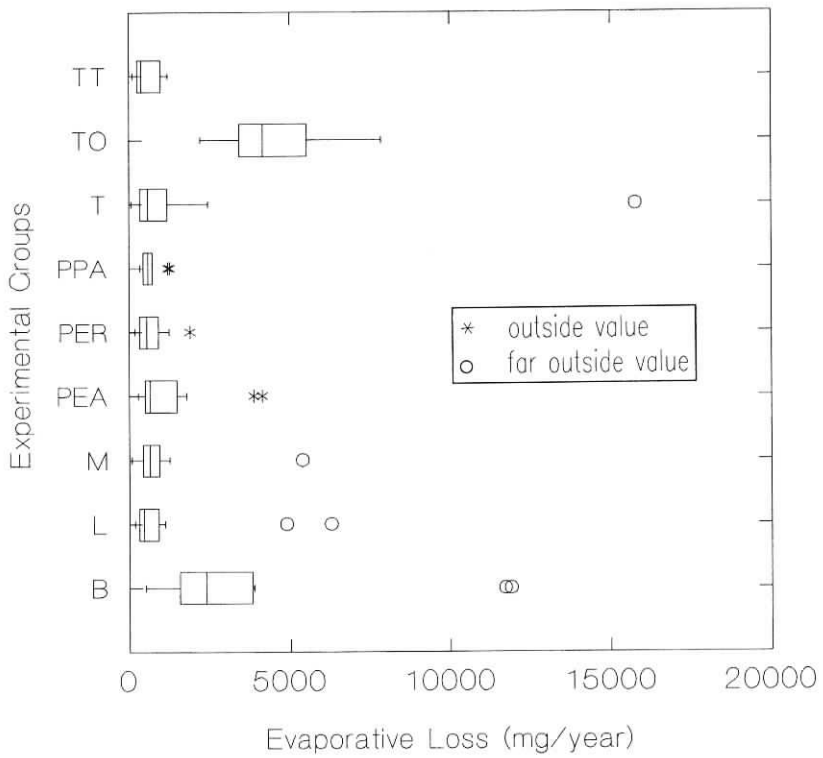

Figure 2. Grouped box plot showing means, 95% confidence intervals and outside values of rates of evaporative loss for each experimental group.

Polyethylene backing, such as that used in groups PER and PEA, is a stable, extremely conformable material (3M, 1989a, b). Stability of the material is an important quality but the conformability was found to be a deterrent after observing, within 6 months, that the tape was drawn into the groove at the jar-lid junction. The resulting displacement of the tape left a sticky residue on the jar while reducing the surface area occupied by the sealing tape on the jar-lid. This action might disrupt the seal over time. The polypropylene-acrylic tape PPA, on the other hand, adhered well and without displacement of the tape into the jar-lid junction. The cause of tape displacement in test group PEA cannot be explained without further investigations. PER tape did not seem to slide into the groove as PEA did, even though it is made of the same polyethylene backing. PER tape, used in the 1975 experiment, had not shown any visible displacement either. The reason why PPA did not react similarly remains unknown. The acrylic adhesive was still very tacky and no residue was noticed around the jar or lid upon removal of the tape after 2 years; however, some yellowing of the polypropylene backing was observed.

PPA and PEA tapes were tested for the acrylic adhesive stability in presence of ethanol. Under a light microscope, a drop of 95% v/v ethanol/water was placed on the acrylic adhesive to check for any sign of reaction. Although the polyethylene acrylic-adhesive tape became opaque within a minute, the adhesive did not dissolve. It remained attached to the polyethylene backing and reacted in the same

way as the control sample which was not exposed to ethanol. This suggests that the polyethylene backing might be somewhat permeable to ethanol. The polypropylene/acrylic-adhesive tape, on the other hand, remained clear and the adhesive seemed unaffected by the alcohol. Samples of PPA and PEA tapes were also tested by immersion in 200 ml of 75% v/v ethanol/water for 120 minutes at room temperature (20°C), and at 50°C. The sample of PEA tape at room temperature showed a small change in adhesive consistency after immersion and became opaque. However, upon drying, the tape regained the same degree of tackiness and its original transparency. The adhesive was visibly softer for the sample immersed at 50°C. It had formed small waves/bumps and stretched considerably when probed. It could also be easily removed from the polyethylene backing when rubbed with a finger. Upon drying, the tape returned to its original transparency but was less tacky than originally. PPA tape seemed unaffected by both tests: the adhesive retained its properties and the polypropylene backing remained clear. This might explain why PPA tape is widely used in the construction industry in Canada to seal insulation sheets because of its wide temperature resistance (−40°C to 105°C) and good ageing properties (3M, 1989c).

In another test, two layers of PPA tape were placed one on top of the other and immersed into 75% v/v ethanol/water at room temperature (20°C) for 120 minutes. The polypropylene remained clear and the adhesive retained its tack. The same test was performed with the PEA tape. The area where the adhesive was sandwiched between the two pieces of polyethylene backing did not become opaque. Its tack also seemed unaffected. These observations suggest that the same type of adhesive can perform differently depending on the type of backing utilized.

Stability of the tape in the presence of ethanol was tested by immersion of two filled jars (one taped with PPA and the other with PEA) into a solution of 70% v/v ethanol/water. After two years, we observed that the PEA tape was no longer adhering to the jar-lid. The adhesive was opaque and detached from the backing. Conversely, PPA tape remained unchanged from its original state. These observations strengthen the good performance ability of PPA tape.

Predicted evaporation over time.—The different test groups were compared by looking at the number of years predicted for 25% evaporative loss to occur. Fluid preservative levels would not normally be allowed to fall below 25%. Njie and Waller (1993) demonstrated that evaporative loss occurs proportionally until it reaches about 50% evaporation. Their experiment showed that alcohol concentration decreases dramatically if preservative fluids evaporate any further.

Confidence intervals were calculated using the *mean* rate of loss based on a volume of 375 ml and the density of 70% ethanol (0.880). The values obtained do not reflect the reality of collection maintenance requirements (Table 3). In most collections, the daily maintenance is dictated by which jars show the most evaporative loss: the worst performing jars are the first to require topping up. The values for 25% evaporation should therefore be calculated using the *maximum* rate of evaporative loss. Although the maximum rate of evaporative loss should be used, it is not statistically possible in our experiment to calculate its precision because it is based on a single value for each group.

A different experimental design is required to realistically demonstrate the evaporative loss occurring in typical fluid-preserved collections. Experiments

Table 3. Number of years predicted to achieve 25% fluid evaporation for each test group.

Test groups	Number of years, predicted for 25% fluid evaporation	95% confidence interval
B	46	28
L	205	93
T	244	229
TT	242	122
TO	21	3
PER	199	86
PEA	130	55
PPA	149	34
M	211	172

should be designed selecting only a large group of the worst performing jars. These jars could then be tested with a variety of tapes, as in our experiment. Statistical results would reflect the realistic effect of the tapes on the rate of evaporative loss. These future experiments may give a good indication as to how many years can elapse before jars must be taken to the laboratory so that their alcohol levels and concentrations can be restored.

Conclusions

Test results illustrate that the application of polypropylene/acrylic-adhesive (PPA) tape reduces evaporative loss significantly when compared to group with lid only (B) or group with lid and liner (L) which are presently used in our collections. The experiment showed that PPA tape is effective and resistant to alcohol and temperature fluctuations. Furthermore, use of PPA tape is not as expensive nor labor intensive as topping up jars, since time spent on routine maintenance can be substantially reduced. With the present economic situation adversely affecting museums, it is more important than ever to see that time spent on collection care is used judiciously. Alternative methods of reducing evaporative loss such as replacing containers with expensive, high efficiency ones might be available, nevertheless, the application of PPA tape is an excellent temporary solution. It can be easily removed when specimens need to be accessed, without leaving sticky adhesive residues. Although the long-term effect of PPA tape on the preservative and specimens has not yet been determined, any effect might be minimal since the tape is placed on the outside of the jar and is not in direct contact with the preservative and specimens. The application of polypropylene/acrylic-adhesive tape around the jar-lid junction provides a cost effective, *practical* method of reducing evaporation in fluid-preserved collections.

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Appendix. Suppliers of equipment and materials used in evaporative loss testing.

Equipment and materials	Suppliers
Aluminum foil adhesive tape	Available at most hardware stores. Also called "Muffler Tape"
Mettler AT400 Balance cat.# 01-910-2	Fisher Scientific 112 Colonnade Road Nepean, Ontario K2E 7L6
PAAR Digital Density Meter DMA 35 cat.# 10-820-5	Fisher Scientific 112 Colonnade Road Nepean, Ontario K2E 7L6
Tapes	
Polyethylene/acrylic-adhesive (PEA) Product #480 Polyethylene/rubber-adhesive (PER) Product #483 Polypropylene/acrylic-adhesive (PPA) Product #5086	3M Canada Inc. P.O. Box 5757 London, Ontario Canada N6A 4T1
Polyethylene liners	Langford Mfg Ltd. R.R. #10, Site 13, Comp. 9 Fredericton, New Brunswick Canada E3B 6H6
Teflon® tape	Available at most hardware or plumbing stores.
375 ml Universal Screw Type Jars	AMPAK Ltd 7544 Côte-de-Liesse Ville St-Laurent, QC. Canada H4T 1E7

NORWEGIAN NATURAL HISTORY MUSEUM COLLECTION COMPUTERIZATION: A FIRST REPORT

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Abstract.—Large collections of natural history specimens are housed in university museums in Norway. To make these more accessible to users, the collections are being computerized for subsequent Internet access. Use of non-professional labor to aid data entry is being partly financed by government employment programs. Supervision and information validation by scientific staff ensure the quality of the resulting database. The combined scientific and non-professional labor has proved useful in a situation where the universities cannot finance sufficient scientific positions.

The largest natural history collections and archives in Norway are housed in the university museums in Bergen, Oslo, Trondheim and Tromsø. These collections contain a diverse pool of raw and processed data on botany, geology, paleontology and zoology, and the museums function as national archives for natural history. Working only from paper-based archives, the retrieval of data is often difficult as the contents are mainly indexed only by a single subject, e.g., species name. A cross-reference list by species name and year of collection would take a long time to prepare. To make the data more readily available to users, the paper-based information has to be converted into an electronic form. This conversion is the main objective of the present project covering the natural history museum collections administrated by the faculties of mathematics and natural sciences at the four Norwegian universities. The aim is to offer access to the computerized collections via an electronic network, e.g., the Internet, thus bypassing geographical and professional borders. Such an information system has several functions:

1. The computer-based registering of specimen data provides for more efficient management of museum specimens and associated information (Williams, 1987). Data is more readily available when stored electronically. Loans can automatically be called back when expired.
2. The computerized information is more accessible to scientists, educators and the public (Hoffmann, 1992; Bearman, 1994).
3. Cross-linked information from different professions becomes of interest to "new" user groups, and can solve new problems and answer questions (e.g., the relationship between geology and plant distribution; the relationship of bedrock chemistry and river chemistry to the distribution of fishes).
4. Use of collections and associated information will increase as the database becomes available to a wider audience. This may reduce exploitation of vulnerable collection sites.

OBJECTIVE

The objective of the present project is to make information in natural history collections electronically available, and to make the use of this information more efficient in the following ways:

1. Computer registration of collections and archives (data entry).—The manual conversion of paper data into computer data is in practical terms, a typing process,

where information from catalogs, archives and specimen labels is entered into an electronic database.

2. *Efficient routines for collection and archive management.*—Automation calls for changes in museum routines such as security strategies (computer file back-up), adaption to and knowledge of computer use, and the handling of health risks (computer screen radiation, office environment). These routines will be monitored during the project period, so that they can be continued beyond the start-up phase.

3. *Access requirements for researchers, government departments and the public.*—Users of the system must have access to unambiguous information. User interface and support will be adapted to each group. A graphical interface (e.g., Microsoft Windows™) will be available where novice users will find fields to fill in to perform queries (Bearman, 1994). Accessibility must, however, be controlled in order to protect collections, vulnerable sample sites, research copyrights and sources of information. Each museum will screen their data sets prior to electronic publication. Locality details (e.g., for birds of prey and unique minerals and fossils) will not be available for occasional users. Such information will only be available for scientists after personal communication with the curator in charge for the specific collection.

4. *Unified data presentation and user interface.*—Efficient information access requires a common user interface. The project's administrative board will define standards in data field catalogs (data modelling) based on professional requirements, existing procedures, and established databases. Linking between different databases will be carried out with common relational fields. The registering itself will be hard- and software-independent as long as compatibility is achieved according to field catalog standards prepared in the project.

Who Are Possible Users of the Natural History Databases?

Natural history museum collections attract different users. First and foremost, the museum curators responsible for the collections will be users of the computerized inventory databases. Curators respond to inquiries from scientists and the public, and they will maintain and update the databases. New acquisitions will be registered, common curatorial processes (e.g., production of labels and annual reports) will be automated as a result of using an electronic cataloging database program, and collection catalogs can be produced for publication. These, and other advantages of a modern approach to collection management, are well outlined in Davis (1994) and references therein.

Developers, planners and politicians may need information regarding protected areas, vulnerable collection sites or endangered plant and animal species when regulating areas for industrial development or housing. Teachers may consult the natural history databases prior to excursions or field work by pupils in special regions.

Public accessibility to the systematized information must be agreed upon with each department involved in order to prevent misuse, as many museum collections contain items of considerable commercial value. Detailed information on special items such as precious minerals, unique fossils and taxidermic mounts of birds of prey will not be included in the open file databases. All updating will take place locally (on local PC-based in-house networks), with the collection manager

monitoring all stages in the registering process. Filtered subsets of information will be copied to open network systems, and made available for external users through the Internet. Basic specimen data will be available: common and scientific name, locality (with varying level of detail), collector, date of collection, and preservation (wet or dry, stuffed, thin section, parts, etc.).

Project Feasibility

The university faculties of mathematics and natural sciences have appointed a governing committee people from the university museums. This committee, together with personnel from university computer departments, defines the aims of the project and coordinates the work. Individual members act as advisors for their respective universities. Guidelines are defined in a project plan.

The primary task is the conversion of data from written labels and catalogs into a machine readable form. Electronic scanning and optical character recognition (OCR) programs are of very limited use as most of the labels and catalogs are handwritten. The size of the collections involved (more than 12 million items) and the rather small number of museum curators working on these collections necessitate additional labor. Government programs have initiated "work training" to help long-term unemployed people. Other cultural institutions have reported good results in using such labor, and the natural history project is also relying on this source. As is always the case, annual budgets at most university museums are rather small, and thus payment of salaries from the Ministry of Employment covers almost all the extra expense of hiring people who are out of work. To date, this has worked successfully at the Paleontological Museum in Oslo, and the Zoological Museum in Bergen. A total of 16 people from this pool of workers are currently carrying out data entry. This figure is expected to increase to approximately 50 within the next few years.

As the data entry will be conducted by people with little professional knowledge, quality control will be carried out by the scientific staff of each collection. A certain degree of validation will be ensured prior to making the collections electronically available.

Before a collection is considered for computerization in the present project, it must meet the criteria defined by the steering committee. Ultimately, all collections are to be computerized, but priority is given to the collections which (1) are actively used in current research projects, (2) contain unique specimens, (3) are considered a national or international scientific source, and (4) lend themselves to computer registration. Each museum may also suggest priority criteria.

A well defined data model has still to be developed, but a data field catalog is in the process of being distributed. An adaption of the information categories recommended by ICOM/CIDOC (1995) is followed in the definition of fields. Those collections included in the project for computer registration will have to follow the standardized core fields in the data field catalog. These include: specimen identification (museum catalog or acquisition number), taxonomic details (animal or plant group, species name), nature of specimen (thin section, stuffed, wet or dry, skeleton, part, etc.), specimen location, locality (country, county, place name with grid reference), stratigraphy (for geological and paleontological items), collector's name and date, and any existing published information. Similar field compositions are documented for (among others) the Birmingham City Museum

Table 1. Estimated number of specimens in the collections, and number of computerized specimens. (*): The zoological collections in Bergen include approximately 50,000,000 osteological specimens from archaeological research in western Norway. These specimens may be computerized as lots, thus reducing the total number of registered items to the corrected sum.

Science	University	Specimens	Computerized
Botany	Bergen	783,000	20,000
	Oslo	1,800,000	100,000
	Tromsø	270,000	25,000
	Trondheim	418,000	20,000
Geology	Bergen	215,000	7,500
	Oslo	250,000	42,000
	Tromsø	25,000	5,000
Paleontology	Bergen	51,000	0
	Oslo	1,740,000	50,000
	Tromsø	5,000	300
Zoology	Bergen(*)	51,000,000	210,000
	Oslo	5,600,000	47,000
	Tromsø	132,500	7,500
	Trondheim	580,000	103,000
SUM		62,869,500	637,300
Corrected sum (*)		12,869,500	

(Seddon, 1982), the Indian Coastal Zone Museum (Mikkelsen, 1986) and the Milwaukee Public Museum (Sumpter, 1991). Certain codes (defined in thesauri) will be used to ease typing of repetitive information, and also to ensure consistency. Both national and international standards (codes), where possible in accordance with International Council of Museums (ICOM) and the International Committee for Documentation (CIDOC), will be used to ensure compatibility with other database management systems. Descriptions of fields to be included in natural history databases can be found in Giles (1995), Waddington and Rudkin (1986), and Williams (1987). International code lists (thesauri) for biological and physiochemical (environmental) terms can be found at the Nordic Code Center (Natural History Museum, Stockholm, Sweden) (Pinborg and Paule, 1990; Österdahl and Österdahl, 1990) or at its ftp-site (ftp://ftp.nrm.se/pub/nrm/it/termlist). Similar terms have also been published by the Documentation Research Group of the Canadian Heritage Information Network (CHIN, 1985).

Collections Included for Registering

Each museum has estimated the size of its collections, but errors are almost certainly involved because of incomplete inventorying (Table 1). Including both unnumbered and fragmented specimens which may be given a catalog number, the collections comprise more than 60 million objects, which may eventually represent more than 12 million valid entries in the system. A trained person, without specific scientific background may register 10,000 to 25,000 specimens a year. The actual typing of data will thus take more than 500 man-years of labor. With a staff of five in each of the twelve museums involved, the project registration would be finished in less than 10 years. This work would need to be supervised by a project scientist or curator attached to each museum. The supervisors, as well as the central administration of the project, will be financed by the uni-

versities. Non-university institutions and directorates may find value in the project and provide additional funding.

Conclusion

As a result of a pilot project started in 1994, collections to be computer registered have been identified and quantified. Local representatives of the scientific and technical staff at the museums have met and discussed this work. Non-professional labor, provided by government employment programs will have to be used in order to meet the aim of this project: to make the natural history collections electronically accessible. Data entry supervision and information validation will be conducted by the scientific staff. Some collections have already been computerized (i.e., all birds in the four zoological museums, and the collection of originals and type specimens at the Paleontological Museum in Oslo), and are in the process of being transferred to a World Wide Web server. Obstacles regarding copyrights, research clauses (confidential reports with time restrictions), lack of labor and space have been overcome during this period. The university faculties of mathematics and natural sciences have agreed to support and continue the project, and it seems possible that all natural history collections at these institutions will be electronically available in a few years.

Acknowledgments

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A STUDY OF THE RESPONSE OF DRY SKIN TISSUE TO WATER SATURATION AND SUBSEQUENT DRYING TREATMENT

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Abstract.—A study was initiated to evaluate the suitability of flood recovery treatments for water-saturated skin tissues associated with bird and mammal collections. A series of untreated, dry skin samples were saturated in water at 20°C, 40°C, and 60°C. Each series was subdivided so that part was dried by various freeze-drying methods. The ambient moisture absorption potential and shrinkage temperature of skin samples were documented for each treatment group and the results were compared to untreated skin samples. Results of these studies support actions taken during and after a specific emergency situation, and suggest that these actions may be appropriate for inclusion in emergency preparedness plans for bird and mammal collections.

In October, 1992, a series of events resulted in a hot-water pipe breaking and flooding the basement collection area of the Texas Cooperative Wildlife Collection, Texas A & M University (Anon., 1992). Flood water reached a depth of 1.1 meters. Portions of all collections (fish, amphibians, reptiles, birds, and mammals) were affected, however, initial concern was primarily for preserved skins and skeletal material of thousands of birds and mammals. At the request of the university, a disaster relief company (Blackmon-Mooring-Steamatic Catastrophe, Inc., Fort Worth, Texas) and four consultants were on-site in a matter of hours to provide recovery assistance. Although most of the water was pumped out of the collection area within 12 hr, many specimens remained in water several hours longer, until water trapped in storage drawers could be removed.

As evacuation and stabilization efforts proceeded, a variety of questions were raised about the best ways to return the collections to normal, and to avoid compounding existing problems through inappropriate activities. The first realization was that this type and magnitude of disaster was essentially new to the natural science community. There were no previous documented experiences involving preserved animal specimens to help direct decision-making and corrective action. As a result, much of the relief effort followed strategies that had been used successfully for flooded libraries (Behrendt, 1981; Koesterer and Geating, 1976; Ruggere and Morse, 1980; Waters, 1979) or collections other than natural science research collections (Gatenby and MacGregor, 1993; Keck, 1972; Schmidt, 1985; Walsh, 1988).

Collection recovery involved a variety of activities working in concert. First, the affected parts of the collection were separated from other parts that had escaped the flood by being above the water level. The affected parts were subdivided according to proposed stabilization treatment. Study skins were removed from the collection area in their original storage cases and placed in large mobile freezers. Freezing was used to mitigate specimen deterioration by fungal and bacteriological action. It was understood that subsequent treatment probably would involve total moisture elimination through a vacuum-drying process.

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Because it was anticipated that wet skeletal material would be at greater risk to mechanical damage by both ice formation during freezing and extreme desiccation during vacuum-drying treatments (Lafontaine and Wood, 1982; Williams, 1991a), all skeletal material was removed before the flooded cases were placed in the mobile freezers. The skeletal material was taken to work areas where standing water was removed from storage containers (boxes and vials) and the specimens were allowed to air-dry.

Because tanning makes skins less susceptible to biodeterioration, but more susceptible to hardening and deformation through static drying, the tanned skins also were moved to other work areas where they could be monitored and manipulated during an air-drying process. Decisions were made not to freeze the tanned skins because of unknown tanning history and concern for reactions of the tanning salts and oils.

Once stabilization of the collections had been achieved, questions were raised about what problems might be expected with bird and mammal study skins subjected to vacuum-based drying treatments. A review of the literature and several bibliographic resources concerning disasters (i.e., Jones, 1986; Meister, 1991; Nelson, 1991) was of questionable value in addressing these questions. Most of the applicable information was based on theory and information extrapolated from case studies, rather than documented experiences or quantitative research. For this reason, a series of studies was initiated to obtain experimental data to provide answers to some of these questions. It is hoped, if and when future disasters involving natural science specimens occur, that the results of these studies might facilitate and substantiate basic decision-making and corrective action.

Methods and Materials

Dry skin of a pronghorn antelope (*Antilocapra americana*) was used to analyze hydration and dehydration treatments. This skin had been removed from the lower legs of a fresh carcass in October 1990. The skin was dried under ambient conditions and stored for about one year in the dark and in a polyethylene bag; thus, the skin samples had a known history of preparation and storage which precluded all other interventive treatments, such as cleaning and fumigation.

In mid-November 1992, the hair was scraped off the skin with a razor blade, and series of 1 × 2 cm samples were cut so that the long-axis followed the long-axis of the leg. Samples were subdivided into 10 groups which received various combinations of hydration and dehydration treatments (Table 1). Because of a limited supply of skin samples, all groups included only 10 samples each, and groups 08, 09, and 10 included only six samples each. Weights were monitored before and after treatments throughout the study, using a Mettler analytical balance, accurate to 0.001 gm. Each skin sample was maintained inside a flint-glass vial. Vials were open during treatments, but sealed with polyethylene caps at all other times. Group 01 was not subjected to any hydration or dehydration treatment.

To hydrate skin samples, distilled water was added to each vial containing a sample. Each group of vials was placed in a water-bath for two hours, where the temperature was controlled by running tap-water and monitored with the use of a centigrade thermometer. Groups 02, 03, and 04 were hydrated at 20°C; groups 05, 06, and 07 were hydrated at 40°C; groups 08, 09, and 10 were hydrated at 60°C (Table 1). Following the hydration treatment, the free water was removed from each vial. To reduce risks of enzymatic action and ice recrystalization, the wet samples were sealed in their respective vials and immediately frozen and maintained in an ultracold freezer (−80°C) until they were removed for one of three drying treatments.

One drying procedure, performed by Blackmon-Mooring-Steamatic Catastrophe, Inc. (BMS; Fort Worth, Texas), allowed frozen samples to thaw from about −3.9°C while under a vacuum. In this situation, ice sublimes and never passes through a liquid state; all moisture is removed by the time thawing is complete. Using a portable unit at the BMS facility, the skin samples were totally dried and thawed in less than two hours.

Table 1. Summary of hydration and dehydration treatments used with groups of skin samples.

		Hydration treatments			Dehydration treatments		
Group	*N*	20°C	40°C	60°C	BMS	CCT	LAB
01	10						
02	10	X			X		
03	10	X				X	
04	10	X					X
05	10		X		X		
06	10		X			X	
07	10		X				X
08	6			X	X		
09	6			X		X	
10	6			X			X

Another drying procedure, performed by Creative Critters Taxidermy (CCT; Lubbock, Texas), maintained frozen samples in a vacuum, at about −23.3°C with a standard freeze-dryer (Northstar Model L-3666). Again, the ice sublimes, but at a colder temperature; thawing is allowed once dehydration is completed. Processing time was one week because other materials were present in the freeze-dryer.

The third drying method was performed under laboratory conditions (LAB). Frozen samples were moved to a standard freezer (−3.9°C). In the freezer, vials containing the water-saturated skin samples were opened and collectively sealed in jars containing oven-desiccated silica gel. In the end, the laboratory efforts became cumbersome and questionable. Primary concerns involved the difficulty of effectively drying samples, protecting samples from condensation during thawing, and ultimately, avoiding the incorporation of extraneous variables that might affect the analyses. Processing was arbitrarily conducted for three weeks.

Following the treatments described the samples were subjected to three forms of analyses. Qualitative analysis involved superficial examination of physical characteristics, specifically alterations of color and shape. Quantitative analysis was performed on moisture absorption potential and shrinkage temperature, as described in the following sections.

Moisture absorption potential.—Weight change of skin samples between different relative humidity (RH) levels provides an indication of the moisture absorption potential. To evaluate the effect of hydration and drying treatments on the moisture absorption potential of skin tissue, all of the samples were subjected to RH extremes of 0% and 75%. Oven-desiccated silica gel was used to lower sample weights to the same levels documented immediately after the freeze-drying treatments. When weights of individual samples were stable for a three-day period, individual weights were recorded. Samples were then acclimated to 75% RH which was established with a saturated sodium chloride solution (Stolow, 1966). When weights stabilized for a three-day period, they were recorded again. The ratio of weight gain was determined for each sample and standard statistics (mean, range, standard deviation, and coefficient of variation) were determined for each group of samples. Statistical analyses of the weight change ratios requires arcsine transformations of the individual values (Sokal and Rohlf, 1969). The transformed values were subjected to a two-factor analysis of variance to compare treatments with one another. To determine if the skin samples had been significantly altered by the treatments, a one-factor analysis of variance was used to compare the untreated samples to the different water temperature treatments of each drying procedure. Statistical analyses were performed with the StatView® SE+ (Abascus Concepts, Inc., 1988) computer program. Significance levels ($P \leq 0.05$) were based on Fisher's probability of least significant difference.

Shrinkage temperature.—Shrinkage temperature (T_s) analysis was used to evaluate the stability of skin tissue subjected to water and drying treatments. T_s provides a means of assessing structural stability of collagen (Haines, 1987). One sample from each treatment group was subjected to ten T_s analyses; analyses for a single sample were spread equally over two days. T_s analyses were performed with a microscope (100× magnification), central processor (Mettler FP90), hot-stage (Mettler FP82HT), and recording printer (Epson FX870). Temperature was increased 2.0°C per minute; sample preparation involved the removal of tissue fragments from the flesh-side of the skin and pre-soaking in distilled water (Williams, 1991b). The initial T_s and final T_s were determined for each analysis.

From these values the temperature range and temperature mid-point were determined. Because mid-point values have been regarded as more indicative of actual T_s (von Hippel and Schleich, 1969), and because they facilitate reporting of results, these values were used to compare groups with one another. The mid-point values were subjected to a two-factor analysis of variance to compare treatments with one another. To determine if the skin samples had been significantly altered by the treatments, a one-factor analysis of variance was used to compare the untreated samples to the different water temperature treatments of each drying procedure. Statistical analyses were performed with the StatView™ SE+ (Abascus Concepts, Inc., 1988) computer program. Significance levels ($P \leq 0.05$) were based on Fisher's probability of least significant difference. These analyses were performed with standard data and log-transformed data to evaluate the uniformity of the analysis of variance for these data.

Results

Visible changes of treatment groups involved color and shape of samples. Samples subjected to 20°C water tended to be paler than the untreated samples, whereas samples subjected to water temperatures above 20°C were not only paler than the untreated samples, but also might include darkened areas (Fig. 1). Some deformation of samples was also noted, particularly among samples subjected to 60°C water (Fig. 1). With regard to drying treatments, a vacuum process (BMS and CCT) generally resulted in samples of uniform shape. Samples dried without a vacuum (LAB) were characteristically darker on the lower half of the samples (Fig. 2).

Moisture absorption potential.—Subtle losses of moisture absorption potential occurred with the treated groups, as compared to the untreated samples (Table 2). Based on group means, the loss of moisture absorption potential ranged from 0.7% to 1.7%. The samples subjected to higher water temperatures tended to have lower moisture absorption potential values.

Because of questions of limited sample sizes for the groups subjected to 60°C water, the two-factor analysis of variance was performed in two stages. The first analysis involved only those groups with sample sizes of 10; the second analysis included the groups with smaller sample sizes. In both analyses, there were no significant differences between treatments (water treatment, $F = 1.853$, $P = 0.179$; drying treatment, $F = 0.132$, $P = 0.876$). However, the one factor analysis of variance showed that samples subjected to water temperatures of 40°C and 60°C experienced a significant loss of moisture absorption potential from the untreated condition (Table 2).

Shrinkage temperature.—Based on group means (Table 3) subtle T_s reductions were noted with treatment groups. The T_s for all groups only ranged from 67.1°C to 65.3°C. The lowest T_s values involved those samples subjected to 40°C water treatment.

The two-factor analysis of variance revealed significant differences between treatments, with most of the differences being attributed to water temperature (water treatment, $F = 26.856$, $P = 0.0001$; drying treatment, $F = 1.351$, $P = 0.265$). The one-factor analysis of variance showed that the 40°C water treatment significantly altered samples from their original condition. Other significantly altered samples included those subjected to 20°C water and laboratory drying, and those subjected to 60°C water and freeze-drying (Table 3). For all analyses, standard data and log-transformed data provided the same results.

Discussion

This study is unique in that it addresses preservation issues associated with an actual emergency situation. While there is an increasing amount of literature con-

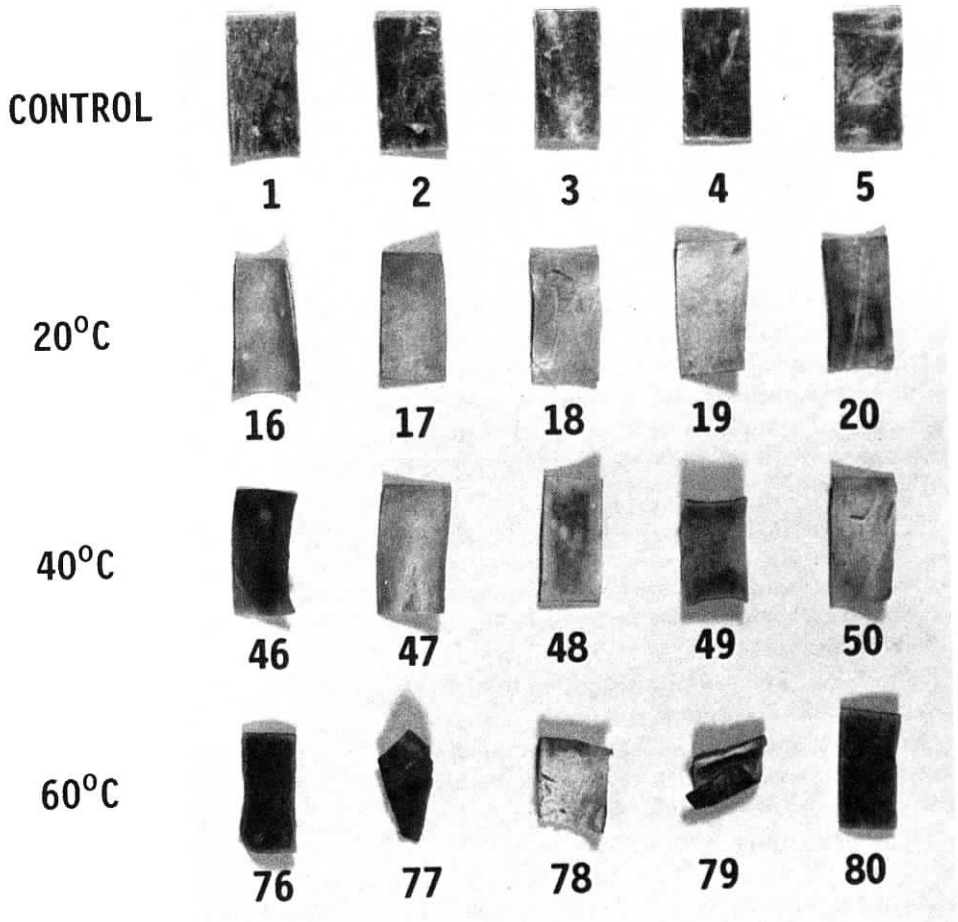

Figure 1.—Photograph of representative dry skin samples showing differences in appearance attributed to hydration at water temperatures of 20°C, 40°C, and 60°C. Numbers identify individual samples.

cerning emergency preparedness and recovery activities, few contributions are based on research intended to evaluate and understand the effects that disasters and salvage procedures have on preservation. Although much of the information in the current study could be anticipated, some new and useful information was obtained.

This study documents that color change, deformation, loss in moisture absorption potential, and T_s reduction of skin tissue may result from various water saturation and drying processes. While any change would be undesirable, it is appropriate to critically evaluate material responses observed in this study before speculating on those that may have occurred as a result of the flood and subsequent recovery activities.

In this study, differences in moisture absorption potential were subtle, but the levels were consistent with previous studies (Kanagy, 1947). It is possible that differences noted could be attributed to soluble components being removed from

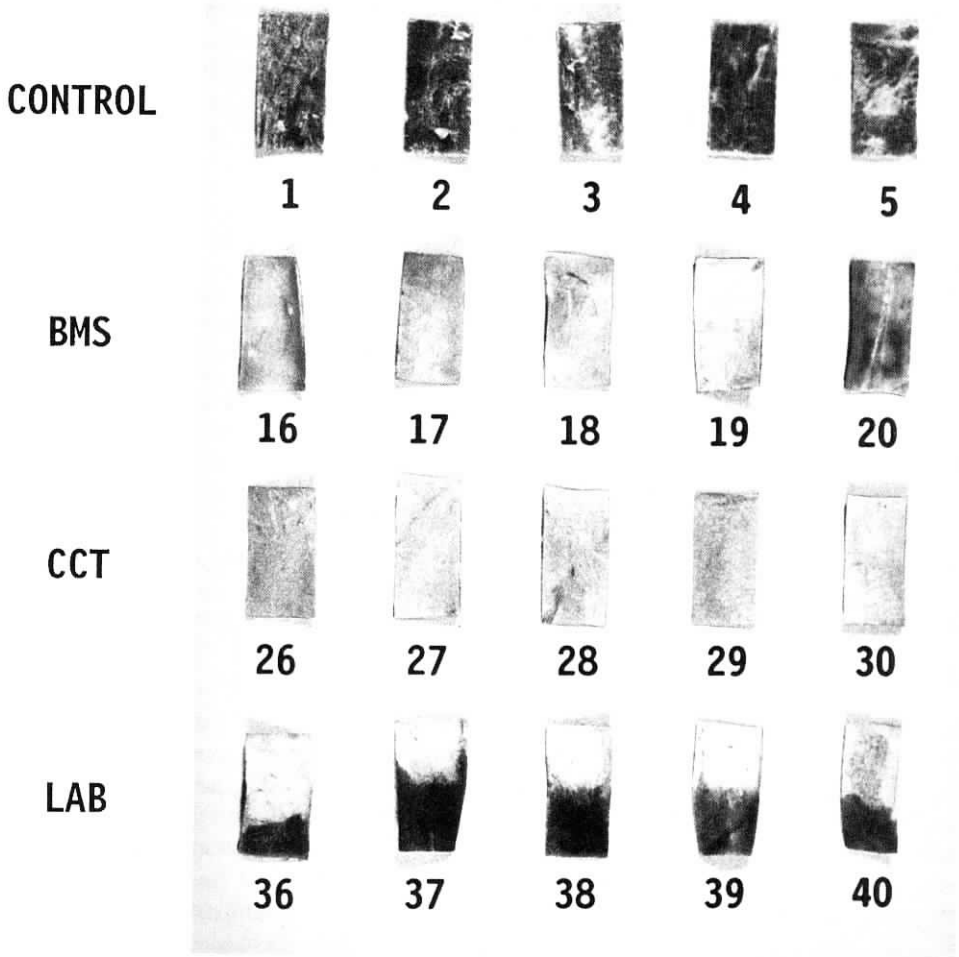

Figure 2.—Photograph of representative dry skin samples (hydrated at water temperatures of 20°C) showing differences in appearance attributed to drying treatments. Numbers identify individual samples.

the skin. As water temperatures increase, further solubilization would be expected with the denaturation of the proteins. The greatest losses in moisture absorption potential were associated with samples subjected to water temperatures above 20°C.

The degradation of skin tissue is a complex and variable process that incorporates interactions of proteins, lipids, and possibly other natural or unnatural components. It is argued that a balance of these components, as expressed by the skin's natural condition, is the preferred state of preservation. An increase in moisture absorption potential would lead to hydrolytic degradation; a decrease in moisture absorption potential can lead to a greater risk of damage because of the reduced ability of skin tissues to respond to stresses caused by environmental change and handling.

This study also demonstrates that some hydration and drying treatments will

Table 2. Standard statistics (sample size, mean, minimum, maximum, standard deviation, and coefficient of variation) of percent moisture weight gain from 0% to 75% relative humidity. Groups are listed in descending order by mean; groups differing significantly from the untreated group (01) in the one factor analysis of variance are indicated with an asterisk (*).

		Percent		
Group	*N*	Mean (min–max) ± STD	CV	Treatment
01	10	19.8 (17.9–20.9) ± 1.0	5.06	No treatment
02	10	19.1 (15.2–20.7) ± 1.5	7.95	20°C H_2O, BMS
03	10	19.0 (17.2–21.8) ± 1.6	8.55	20°C H_2O, CCT
04	10	18.8 (15.7–20.9) ± 1.4	7.61	20°C H_2O, LAB
05*	10	18.5 (16.0–21.1) ± 1.3	7.35	40°C H_2O, BMS
06*	10	18.5 (16.3–20.7) ± 1.4	7.48	40°C H_2O, CCT
10*	6	18.4 (16.7–20.8) ± 1.3	7.19	60°C H_2O, LAB
07*	10	18.3 (17.0–20.5) ± 1.2	6.39	40°C H_2O, LAB
08*	6	18.1 (16.8–20.0) ± 1.1	5.93	60°C H_2O, BMS
09*	6	18.1 (16.8–20.0) ± 1.1	5.93	60°C H_2O, CCT

reduce the T_s of collagenous tissues. Such a reduction is important because it represents a general decrease in stability and an increase in susceptibility to other degradation processes. However, it is important to note that the T_s means for all groups fell within a two degree range.

The fact that the T_s of skin tissue subjected to 40°C was lower than that subjected to 60°C is noteworthy. High temperatures contribute to collagen degradation because of protein denaturation and the increased loss of hydrogen bonding that holds the collagen strands together. Much of the collagen degradation has already occurred when temperatures reach 60°C (Young, 1990), thus leaving only the most resistant components in place for the phase transition (Gustavson, 1956). In this study, the resistant parts were expressed with the mid-point T_s (a result of a shorter T_s range with higher values) being closer to the T_s of the untreated samples. The only exception to this pattern was observed with the samples subjected to 20°C water and dried under laboratory conditions. In this situation, the drying treatment apparently had a negative effect on the collagen stability.

Table 3. Standard statistics (sample size, mean, minimum, maximum, standard deviation, and coefficient of variation) of mid-point shrinkage temperature values. Groups are listed in descending order by mean; groups differing significantly from the untreated group (01) in the one factor analysis of variance are indicated with an asterisk (*).

Group	*N*	Mean (min–max) ± STD	CV	Treatment
01	10	67.1 (66.7–67.3) ± 0.2	0.35	No treatment
02	10	67.0 (65.3–68.3) ± 0.9	1.36	20°C H_2O, BMS
03	10	66.9 (66.5–67.4) ± 0.3	0.40	20°C H_2O, CCT
10	10	66.8 (66.2–67.3) ± 0.4	0.61	60°C H_2O, LAB
09*	10	66.6 (66.1–67.3) ± 0.3	0.51	60°C H_2O, CCT
04*	10	66.3 (66.0–66.6) ± 0.5	0.67	20°C H_2O, LAB
08*	10	66.3 (65.6–67.1) ± 0.2	0.31	60°C H_2O, BMS
07*	10	66.2 (65.1–67.3) ± 0.7	1.10	40°C H_2O, LAB
06*	10	65.7 (64.9–66.5) ± 0.6	0.91	40°C H_2O, CCT
05*	10	65.3 (64.7–66.5) ± 0.7	1.00	40°C H_2O, BMS

It is important to realize that this study suggests that under certain conditions degradation can be expected, regardless of the quality of skin tissues. It is conceivable that the magnitude of degradation may be greater for tissues that have already been compromised by other factors, such as fumigants, light, acidic storage conditions, and natural defects.

It is important also to recognize the differences between the experimental design and the actual emergency situation at Texas A & M University. For instance, the collection was under water several hours longer than test samples, the flood water contained contaminants (i.e., dirt and alcohol), and the specimens were subjected to varying temperatures somewhere between 20°C and 60°C as the water cooled. It is possible that the study skins were damaged more than the skin samples used in this study (excluding the 60°C water treatments).

Any emergency situation can result in a variety of decisions and actions that might exacerbate preservation problems, and there will always be circumstances that could have been handled better if different decisions or actions had taken place. When such situations occur, it is important to realize that recovery treatments often will be necessary to reach a satisfactory level of stabilization. It is conceivable that emergency recovery efforts might involve a variety of reasonably inexpensive "in-house" remedies. This study demonstrates that such actions can easily lead to undesirable results. For example, skin samples treated under lab conditions, rather than with equipment and expertise suited for the situation, experienced more damage. Therefore, it is recommended that such "in-house" remedies be minimized or avoided completely.

Conclusions

It is assumed that the specimens affected by the Texas A & M University flood were damaged, and that the damage could have exceeded levels demonstrated in parts of this study. In all likelihood, most of the damage was caused by the flood itself, and possibly by handling during the evacuation process. However, any damage to the moisture absorption potential or the collagen stability of the skin tissues caused by the freeze-drying treatments, is regarded as not significant. Furthermore, such treatment is probably unavoidable once the specimens have been subjected to flood conditions.

Future use of the collection will be a determining factor as to whether the flood and subsequent recovery seriously affected the research potential of individual specimens. For traditional uses, it is likely that the collection still has considerable value. However, uses involving biochemical and genetic analyses may be more limited and should be pursued cautiously.

Considering the nature and magnitude of the problem, as well as the need for immediate response, the bird and mammal collections at Texas A & M University recovered exceptionally well from the flood in 1992. This study supports the decisions and corrective actions that were taken to mitigate the effects of this unfortunate event.

Acknowledgments

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PHYSICAL CHEMICAL PROPERTIES OF PRESERVATIVE SOLUTIONS—I. ETHANOL-WATER SOLUTIONS

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Abstract.—This paper presents data on the physical-chemical properties of ethanol-water solutions over the concentration range of 0–100%. It describes, tabulates, and graphs the relations between volume percent, weight percent and mole fraction as measures of the concentration of ethanol-water solutions. Data are presented on partial and total vapor pressures, vapor phase concentration, equilibrium relative humidity, water activity, osmotic pressure, freezing point, boiling point, refractive index, flash point, the dielectric constant, and antiseptic effect as functions of concentration.

Ethanol has been used as a fluid preservative for more than 300 years (Simmons, 1993) and is now probably the most common fluid-preservative for whole-animal biological specimens (Simmons, 1987). It is also a chemical that is widely used commercially, both for consumption as beverages and for industrial purposes as a solvent and chemical intermediary. The annual world production and use of ethanol is in the range of billions of liters (Phaff and Mrak, 1971). For many commercial applications, reviews of the pertinent physical-chemical properties are available (e.g., Mellan, 1977).

This paper presents a compilation of those physical-chemical properties of ethanol-water solutions that are of particular interest to workers in the field of biological specimen preservation. The inspiration to bring this information together has come from the frustration that we have felt in spending hours to find or compute a value for a property at a particular concentration because the needed information was not available in the required form in an accessible source.

Data are presented for a number of properties in three forms: tabular, keyed on volume percent ethanol; graphical, plotted as a function of volume percent ethanol; and, where it was thought to be helpful, mathematical, in the form of empirical equations that are readily programmable. Readers should bear in mind that this is not a critical review of the literature on these properties. We have, rather, in most cases used the most recent or most complete studies available to us. The literature pool considered was generated primarily by a search of Chemical Abstracts, by consulting the references found in those papers, by searches of selected library catalogues, and by reference to our own collections of offprints. The Chemical Abstracts search was for "ethanol" or "ethyl alcohol" combined with "water" and "thermodynamics" and "English" and not "osmosis" over the years 1967 to 1985. Researchers who have a need for the most accurate data possible should, at a minimum, consult the primary references listed with each property below.

The data in this paper are applicable to solutions of pure, undenatured ethanol. Denatured ethanol for preserving solutions may contain one or more of a large number of possible additives. Each of the many Specially Denatured Alcohol

Table 1. Formulations of Specially Denatured Alcohols authorized for use as preserving solutions by the United States Bureau of Alcohol, Tobacco and Firearms. Values given are the amounts of each additive per hundred US gallons of alcohol (from USI Chemicals, 1981).

Formulation	Additive	Added to every 100 gallons
SDA 1-1	methanol	4 gallons
	denatonium benzoate, N.F.	⅛ avdp. ounce
SDA 3A	methanol	5 gallons
SDA 12A-1	benzene	5 gallons
SDA 12A-3	toluene	5 gallons
SDA 13A	ethyl ether	10 gallons
SDA 22	formaldehyde solution, U.S.P.	10 gallons
SDA 23A	acetone, N.F.	8 gallons
SDA 30	methanol	10 gallons
SDA 32	ethyl ether	5 gallons
SDA 37	eucalyptol, U.S.P.	45 fluid ounces
	thymol, N.F.	30 avdp. ounces
	menthol, U.S.P.	20 avdp. ounces
SDA 38B	choice of one or more of a wide range of authorized denaturants	10 pounds
SDA 42 (1)	potassium iodide, U.S.P.	80 grams
	red mercuric iodide, N.F.	109 grams
SDA 42 (2)	thimerosal, N.F.	95 grams
SDA 42 (3)	one of several possible phenyl mercuric salts	76 grams
SDA 44	n-butyl alcohol	10 gallons

(SDA) formulations that are authorized by the United States Bureau of Alcohol, Tobacco and Firearms, Department of the Treasury, Part 212 of Title 27, Code of Federal Regulations, Code no. 430 for use as preserving solutions will have properties that differ from each other and from those listed here for pure ethanol solutions. The formulations for most of these SDA's are given in Table 1. In Canada, the range of SDA's authorized for use as a preservative fluid is much more limited. Information on their formulation is available from Revenue Canada (1995).

In relation to the occurrence of mercury in natural science collections (Hawks and Von Endt, 1990), it is interesting to note that mercuric salts are employed in two of these SDA formulations. It is expected, however, that at this time, only undenatured alcohol solutions are being used in museum collections.

Properties of Ethanol—Water Solutions

Volume percent (%v/v).—This paper presents all data as a function of volume percent ethanol. This is for the convenience of the anticipated users who, almost exclusively, formulate and express the concentration of their solutions in this way.

Volume percent is the percent ratio, at a specific temperature, of the volume of anhydrous ethanol contained in a given volume of solution to that given volume. Hence:

$$\%\text{v/v} = \left(\frac{\text{VOLUME}_{ethanol}}{\text{VOLUME}_{solution}}\right) \cdot 100$$

Table 2. Volume percent concentration of ethanol-water solutions at 15, 20, and 25°C as a function of density (from Revenue Canada, 1980).

Density (g/mL)	% v/v ethanol			Density (g/mL)	% v/v ethanol		
	15°C	20°C	25°C		15°C	20°C	25°C
				0.880	73.8	72.2	70.6
				0.882	73.0	71.4	69.8
				0.884	72.2	70.6	69.0
0.786			99.8	0.886	71.4	69.8	68.2
0.788			99.4	0.888	70.6	69.0	67.4
0.790		99.9	99.0	0.890	69.8	68.2	66.5
0.792		99.5	98.6	0.892	69.0	67.4	65.7
0.794	99.9	99.1	98.2	0.894	68.2	66.5	64.8
0.796	99.5	98.6	97.7	0.896	67.4	65.7	64.0
0.798	99.1	98.2	97.2	0.898	66.5	64.9	63.1
0.800	98.7	97.8	96.8	0.900	65.7	64.0	62.3
0.802	98.2	97.3	96.3	0.902	64.8	63.1	61.4
0.804	97.8	96.8	95.8	0.904	64.0	62.3	60.5
0.806	97.3	96.3	95.3	0.906	63.1	61.4	59.6
0.808	96.9	95.9	94.8	0.908	62.2	60.5	58.7
0.810	96.4	95.4	94.2	0.910	61.3	59.6	57.8
0.812	95.9	94.8	93.7	0.912	60.4	58.7	56.9
0.814	95.4	94.3	93.2	0.914	59.5	57.8	56.0
0.816	94.9	93.8	92.6	0.916	58.6	56.9	55.1
0.818	94.4	93.2	92.0	0.918	57.7	55.9	54.1
0.820	93.8	92.7	91.5	0.920	56.8	55.0	53.2
0.822	93.3	92.1	90.9	0.922	55.8	54.0	52.2
0.824	92.7	91.5	90.3	0.924	54.9	53.1	51.2
0.826	92.2	91.0	89.7	0.926	53.9	52.1	50.2
0.828	91.6	90.4	89.1	0.928	52.9	51.1	49.2
0.830	91.0	89.8	88.4	0.930	51.9	50.1	48.2
0.832	90.4	89.1	87.8	0.932	50.9	49.0	47.2
0.834	89.8	88.5	87.2	0.934	49.9	48.0	46.1
0.836	89.2	87.9	86.5	0.936	48.8	46.9	45.0
0.838	88.6	87.2	85.9	0.938	47.8	45.9	43.9
0.840	88.0	86.6	85.2	0.940	46.7	44.7	42.8
0.842	87.3	85.9	84.5	0.942	45.6	43.6	41.6
0.844	86.7	85.3	83.9	0.944	44.4	42.4	40.5
0.846	86.0	84.6	83.2	0.946	43.2	41.3	39.3
0.848	85.3	83.9	82.5	0.948	42.0	40.0	38.0
0.850	84.7	83.3	81.8	0.950	40.8	38.8	36.7
0.852	84.0	82.6	81.1	0.952	39.5	37.5	35.4
0.854	83.3	81.9	80.4	0.954	38.1	36.1	34.1
0.856	82.6	81.2	79.7	0.956	36.8	34.7	32.7
0.858	81.9	80.5	78.9	0.958	35.3	33.3	31.2
0.860	81.2	79.7	78.2	0.960	33.8	31.7	29.7
0.862	80.5	79.0	77.5	0.962	32.2	30.2	28.2
0.864	79.8	78.3	76.7	0.964	30.5	28.5	26.6
0.866	79.1	77.5	76.0	0.966	28.8	26.8	24.9
0.868	78.3	76.8	75.2	0.968	27.0	25.1	23.3
0.870	77.6	76.1	74.5	0.970	25.1	23.3	21.5
0.872	76.8	75.3	73.7	0.972	23.1	21.4	19.8
0.874	76.1	74.5	73.0	0.974	21.1	19.6	18.1
0.876	75.3	73.8	72.2	0.976	19.1	17.7	16.3
0.878	74.6	73.0	71.4	0.978	17.1	15.9	14.6

Table 2. Continued.

Density (g/mL)	% v/v ethanol 15°C	20°C	25°C	Density (g/mL)	% v/v ethanol 15°C	20°C	25°C
0.980	15.2	14.1	12.9	0.990	6.4	5.8	5.0
0.982	13.3	12.3	11.2	0.992	4.9	4.3	3.5
0.984	11.5	10.6	9.6	0.994	3.4	2.9	2.1
0.986	9.7	8.9	8.0	0.996	2.0	1.5	0.8
0.988	8.0	7.3	6.5	0.998	0.6	0.1	

The units used to measure volumes are irrelevant provided the same units are used for both the ethanol and the solution. It is worthwhile noting that VOLUME$_{solution}$ is not equal to the sum of the volume of ethanol and the volume of water used to make the solution. This is due to a contraction in volume that accompanies dissolution. For solutions in the range of 40 to 90%v/v the error that would result from neglecting this effect when preparing solutions is approximately 2%*v/v*. For example, at 20°C, 70.00 mL ethanol and 30.00 mL of water combine to give 96.84 mL of solution with a concentration of 72.28%*v/v*. Avoidance of this error in preparing stock solutions requires final adjustment of solution volumes after complete mixing and dissolution. Measurement of final concentration indicated by density is the best means of assurance that a correct concentration has been obtained.

Volume percent is also a function of both concentration and temperature. Without specification of the temperature, it is not a definitive specification of the concentration. Although neglect of this fact does not lead to significant errors in all but the most exacting work, we feel that it is wise to first recognize the fact and then dismiss it as insignificant rather than ignore the fact completely.

The uncertainty of volume percent as an expression of concentration derives from the fact that pure ethanol and ethanol-water solutions have different coefficients of thermal expansion. As an example, a 70.00%v/v solution at 20°C is equivalent to a 69.78%v/v solution at 0°C and a 70.18%v/v solution at 40°C. This inequivalence of volume percent concentrations is nonexistent for 0 and 100%v/v solutions and is greatest for solutions near 50%*v/v*. Even where it is greatest, however, the effect is only equivalent to about 0.014%v/v/°C.

These discrepancies are minimal when temperatures considered are around normal room temperatures of 15 to 30°C. For this reason the temperature effect on volume percent concentration will henceforth be ignored in this paper. All expressions of volume percent in this paper, when not otherwise specified, are volume percent at 20°C, one of the standard temperatures for alcoholometry. In cases where exact determinations of the quantity of ethanol present are required (e.g., for taxation), great care must be taken to ensure that all volume percent concentrations are referenced to a standard temperature.

Proof.—Proof is a measure of ethanol concentration used for taxation purposes and is defined as twice the volume percent of ethanol in a sample measured at 15.56°C (60°F):

$$\text{Proof} = 2\cdot(\%\text{v/v}_{15.56^\circ\text{C}})$$

Density (ρ).—Density is the mass of a substance contained in a given volume

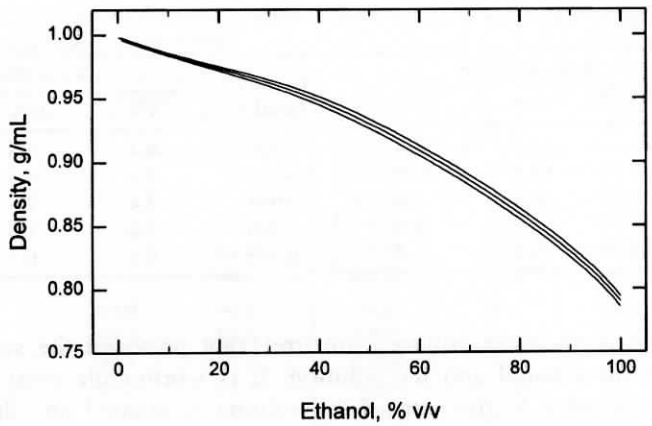

Figure 1. Density of ethanol-water solutions at: 15°C, upper curve; 20°C, middle curve; and 25°C, lower curve.

at a given temperature. The units used here are grams per millilitre (g/mL). Density measurements are the primary method of determining the concentration of ethanol-water solutions and are an essential bridge to the other expressions of concentration discussed later. Densities of volume percent solutions at several temperatures are given in Table 2 and shown in Figure 1. There is no simple equation to relate density to volume percent ethanol. Consequently, tables are generally used to relate density to percent ethanol and vice versa. Fortunately, detailed tables are available in Canada in the Revenue Canada, Customs and Excise "Canadian Alcoholometric Tables" (Revenue Canada, 1980) and in the United States in the Department of the Treasury, Bureau of Alcohol, Tobacco and Firearms "Gauging Manual" (Internal Revenue Service, 1970). In addition, one of us (TS) has designed, and will send on request, a computer program to calculate volume percent ethanol from temperature and density data.

Specific gravity is equivalent to the density of the solution divided by the density of water at temperatures which must be specified for both the solution and the water. The use of specific gravity as a measure is discouraged in favour of the simpler measure, density.

Weight percent (%w/w).—Most data available on the properties of ethanol-water solutions are expressed as a function of weight percent concentration. Consequently, it is important to be able to convert between volume and weight percent expressions of concentration. Values of weight percent concentration that correspond to volume percent concentrations are given in Table 3 and shown in Figure 2. Weight percent and volume percent are identical at 0 and 100% and show a maximum deviation (at 25°C) of 8% at 61%v/v. Volume percent and weight percent are related by the following simple formulae:

$$\%\text{v/v} = \%\text{w/w} \cdot \left(\frac{\rho_{solution}}{\rho_{ethanol}} \right)$$

and

Table 3. Properties of ethanol-water solutions at 25°C.

% v/v	% w/w	X_{water}	$X_{ethanol}$	P_{water}	$P_{ethanol}$	P_{total}	Vapor as % v/v	Vapor-Sol'n as % v/v	%RH	Flash Point, °C	ϵ
0	0.00	1.000	0.000	3.17	0.00	3.17	0.00	0.00	100.0		78.47
2	1.58	0.994	0.006	3.15	0.18	3.33	15.63	13.63	99.5		77.60
4	3.17	0.987	0.013	3.13	0.36	3.50	27.98	23.98	98.9		76.72
6	4.77	0.981	0.019	3.11	0.55	3.66	37.48	31.48	98.2	60.6	75.83
8	6.37	0.974	0.026	3.09	0.72	3.82	44.67	36.67	97.6	55.6	74.93
10	7.98	0.967	0.033	3.07	0.90	3.98	50.60	40.60	97.0	51.5	74.01
12	9.60	0.960	0.040	3.05	1.09	4.14	55.51	43.51	96.3	48.1	73.09
14	11.23	0.953	0.047	3.03	1.26	4.29	59.43	45.43	95.7	45.2	72.16
16	12.87	0.945	0.055	3.01	1.43	4.44	62.80	46.80	95.0	42.7	71.21
18	14.51	0.938	0.062	2.99	1.60	4.59	65.70	47.70	94.3	40.5	70.25
20	16.16	0.930	0.070	2.97	1.77	4.74	68.21	48.21	93.7	38.6	69.29
22	17.82	0.922	0.078	2.95	1.94	4.89	70.34	48.34	93.0	36.9	68.31
24	19.49	0.914	0.086	2.93	2.10	5.03	72.22	48.22	92.4	35.4	67.32
26	21.16	0.905	0.095	2.91	2.26	5.17	73.88	47.88	91.7	34.0	66.32
28	22.85	0.896	0.104	2.88	2.42	5.30	75.34	47.34	91.0	32.8	65.32
30	24.55	0.887	0.113	2.86	2.57	5.43	76.65	46.65	90.4	31.7	64.30
32	26.26	0.878	0.122	2.84	2.72	5.56	77.82	45.82	89.7	30.8	63.27
34	27.98	0.868	0.132	2.82	2.86	5.68	78.85	44.85	89.1	29.9	62.23
36	29.72	0.858	0.142	2.80	2.99	5.80	79.79	43.79	88.4	29.1	61.19
38	31.47	0.848	0.152	2.78	3.12	5.91	80.62	42.62	87.8	28.4	60.13
40	33.24	0.837	0.163	2.76	3.25	6.01	81.37	41.37	87.1	27.8	59.06
42	35.03	0.826	0.174	2.74	3.37	6.11	82.04	40.04	86.5	27.2	57.99
44	36.84	0.814	0.186	2.72	3.48	6.20	82.65	38.65	85.9	26.7	56.90
46	38.66	0.802	0.198	2.70	3.58	6.29	83.20	37.20	85.3	26.2	55.81
48	40.51	0.790	0.210	2.68	3.68	6.37	83.69	35.69	84.7	25.7	54.71
50	42.38	0.777	0.223	2.67	3.77	6.44	84.14	34.14	84.1	25.3	53.60
52	44.27	0.763	0.237	2.65	3.86	6.51	84.55	32.55	83.6	25.0	52.48
54	46.18	0.749	0.251	2.63	3.94	6.57	84.92	30.92	83.0	24.6	51.35
56	48.11	0.734	0.266	2.61	4.02	6.63	85.26	29.26	82.5	24.3	50.22
58	50.07	0.718	0.282	2.60	4.09	6.69	85.57	27.57	82.0	23.9	49.08
60	52.05	0.702	0.298	2.58	4.16	6.74	85.87	25.87	81.4	23.6	47.93
62	54.06	0.685	0.315	2.56	4.22	6.78	86.16	24.16	80.9	23.3	46.77
64	56.09	0.667	0.333	2.54	4.29	6.83	86.45	22.45	80.3	23.0	45.61
66	58.15	0.648	0.352	2.52	4.35	6.88	86.74	20.74	79.6	22.7	44.44
68	60.24	0.628	0.372	2.50	4.42	6.93	87.03	19.03	78.9	22.4	43.27
70	62.35	0.607	0.393	2.47	4.50	6.97	87.35	17.35	78.1	22.1	42.09
72	64.51	0.585	0.415	2.45	4.58	7.03	87.69	15.69	77.2	21.8	40.90
74	66.69	0.561	0.439	2.41	4.67	7.08	88.07	14.07	76.1	21.5	39.71
76	68.91	0.536	0.464	2.37	4.77	7.14	88.49	12.49	74.8	21.2	38.52
78	71.17	0.509	0.491	2.32	4.88	7.20	88.94	10.94	73.3	20.8	37.32
80	73.46	0.480	0.520	2.26	5.00	7.26	89.45	9.45	71.5	20.4	36.12
82	75.80	0.449	0.551	2.20	5.14	7.33	90.01	8.01	69.3	20.1	34.92
84	78.18	0.416	0.584	2.11	5.29	7.40	90.63	6.63	66.7	19.6	33.72
86	80.62	0.381	0.619	2.02	5.45	7.47	91.30	5.30	63.6	19.2	32.52
88	83.11	0.342	0.658	1.90	5.64	7.54	92.05	4.05	59.9	18.8	31.32
90	85.66	0.300	0.700	1.75	5.86	7.61	92.91	2.91	55.4	18.3	30.12
92	88.30	0.253	0.747	1.57	6.11	7.68	93.90	1.90	49.5	17.7	28.93
94	91.02	0.201	0.799	1.33	6.42	7.75	95.08	1.08	41.8	17.2	27.75
96	93.86	0.143	0.857	1.00	6.81	7.81	96.50	0.50	31.5	16.5	26.57
98	96.83	0.077	0.923	0.56	7.30	7.86	98.16	0.16	17.7	15.5	25.41
100	100	0.000	1.000	0.00	7.89	7.89	100		0.0	12.0	24.27

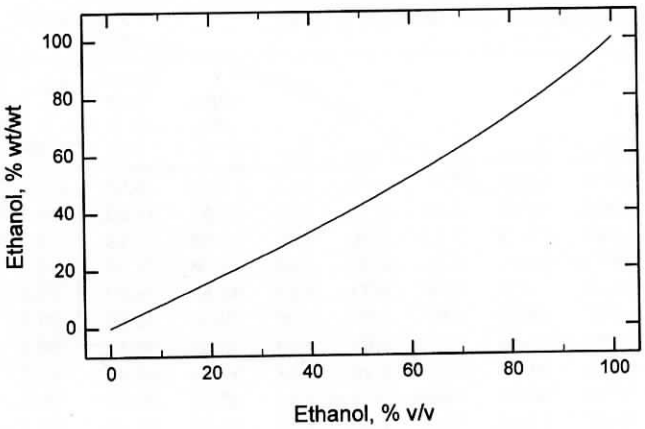

Figure 2. Weight percent concentration as a function of volume percent concentration for ethanol-water solutions at 25°C.

$$\%w/w = \%v/v \cdot \left(\frac{\rho_{ethanol}}{\rho_{solution}}\right)$$

where:

$\rho_{ethanol}$ = density of pure ethanol
$\rho_{solution}$ = density of the solution
%v/v = volume percent of ethanol
%w/w = weight percent of ethanol

It is essential to know $\rho_{solution}$ as a function of %w/w in order to convert from %w/w. Similarly, it is essential to know $\rho_{solution}$ as a function of %v/v in order to convert from %v/v. The former is available in most compendia of chemical data while the latter is found in tables of densities referenced in the above section on density.

Mole fraction ($X_{ethanol}$, X_{water}).—Mole fraction is a dimensionless expression of concentration. It is the number of moles of solute divided by the total number of moles in the solution. Since a mole is simply a large but definite ($6.02 \cdot 10^{23}$) number of molecules, mole fraction is also the same as the number of molecules of solute divided by the total number of molecules comprising the solution. For example, if a solution contains 2 molecules of ethanol for every 8 molecules of water the mole fraction of ethanol would be = 2/(2 + 8) = 0.2 and the mole fraction of water would be 8/(2 + 8) = 0.8.

Values of mole fraction corresponding to volume percent concentrations are given in Table 3 and shown in Figure 3. It is clear from Figure 3 that, while mole fraction and volume percent are equivalent at the endpoints (0 at 0%v/v and 1 at 100%v/v), they differ widely in the midrange. Many properties of ethanol-water solutions are much more nearly linear when plotted against mole fraction than they are when plotted against volume percent since they depend more on the relative number of molecules present than on the volume in solution that those molecules occupy. For this reason many solution properties, when plotted against volume percent, will result in a curve that is similar in form to one of the two curves in Figure 3.

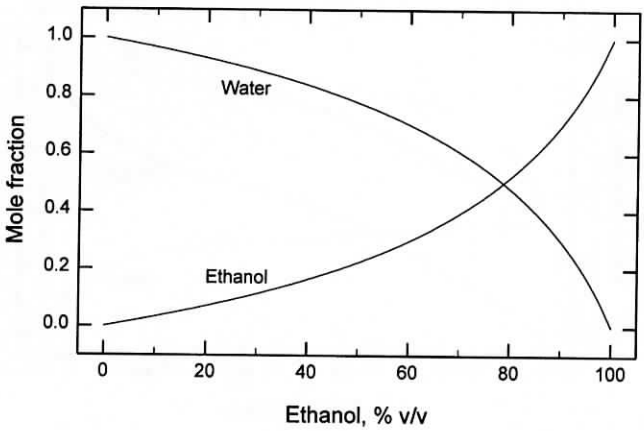

Figure 3. Mole fraction of ethanol, and of water, as a function of volume percent concentration for ethanol-water solutions at 25°C.

It is also interesting to note that the commonly used 70 to 75%v/v solutions of ethanol contain approximately equal numbers of molecules of ethanol and water. Volume percent and mole fraction are related by the following formulae:

$$\%\text{v/v} = 100 \cdot \left(\frac{\rho_{solution}}{\rho_{ethanol}}\right) \cdot \left(\frac{X_{ethanol} \cdot MW_{ethanol}}{X_{ethanol} \cdot MW_{ethanol} + (1 - X_{ethanol}) \cdot MW_{water}}\right)$$

and

$$X_{ethanol} = \frac{\left(\frac{\Phi}{MW_{ethanol}}\right)}{\left(\frac{\Phi}{MW_{ethanol}}\right) + \left(\frac{(1 - \Phi)}{MW_{water}}\right)}$$

where:

$$\Phi = \left(\frac{\%\text{v/v} \cdot \rho_{ethanol}}{100 \cdot \rho_{solution}}\right)$$

and

$MW_{ethanol}$ = Molecular weight of ethanol (46.07 g·mol^{-1})
MW_{water} = Molecular weight of water (18.015 g·mol^{-1})

Vapor pressure (P_{total}, $P_{ethanol}$, P_{water}).—All collection managers and conservators who deal with fluid-preserved collections are familiar with the fact that ethanol solutions tend to evaporate. Vapor pressure is a measure of the tendency of components to escape from a solution to the gas phase. The vapor pressures (at 25°C) of ethanol, of water and of the two combined as a function of volume percent ethanol are given in Table 3 and shown in Figure 4. These data are calculated from the thermodynamic values tabulated by Larkin and Pemberton (1976).

Vapor phase concentration.—A quantity that is closely related to the vapor pressures of the components is the vapor phase concentration. This quantity has considerable importance to managers of fluid-preserved collections since it ap-

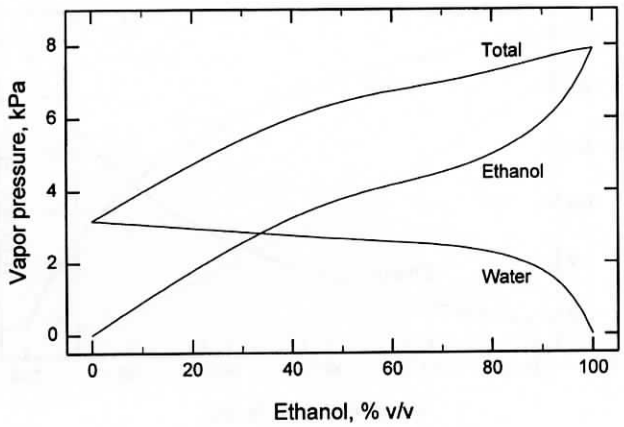

Figure 4. Vapor pressures of water, of ethanol, and of both totaled, as functions of volume percent concentration at 25°C.

proximately quantifies the tendency for solutions to become more dilute in ethanol as they evaporate. It is only an approximate quantification because, to be exact, the effect of ambient relative humidity and certain other factors beyond the scope of this paper must also be considered. Although the relation of vapor-phase to liquid-phase concentration is normally expressed by a comparison of their respective mole fractions, the data are presented here in a manner that is thought to be more useful to fluid-preservative workers.

Here, the vapor phase concentration is expressed as the concentration of the solution that would be formed by condensing the vapor. This quantity is tabulated in Table 3 and shown graphically in Figure 5. Also given in Table 3 and shown in Figure 5 is the difference between vapor and solution phase concentrations. These data indicate the relative amounts, expressed in solution volume percent, of ethanol and water that leave a solution when it evaporates into an ambient

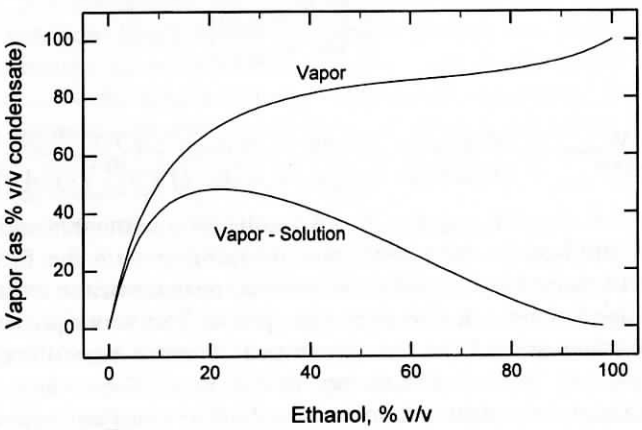

Figure 5. Vapor phase concentration, expressed as volume percent concentration of the solution which would condense from the vapor, and difference between vapor and solution concentrations, as functions of volume percent concentration of the solution at 25°C.

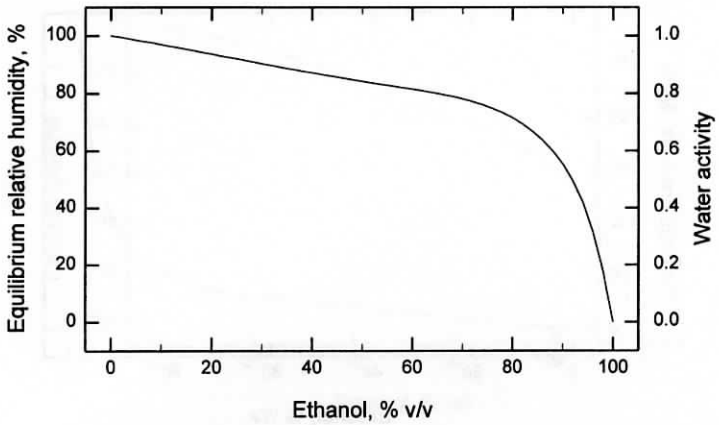

Figure 6. Equilibrium relative humidity and water activity as functions of volume percent concentration of the solution at 25°C.

atmosphere with zero percent relative humidity (RH). For example, a solution of 70%v/v evaporating into air at 0%RH would give off vapor consisting of 87%v/v ethanol, when expressed as %v/v ethanol in the condensed vapor. 87%v/v ethanol was the minimum concentration of stock solution for maintaining preservative levels that was recommended to the Division of Fishes of the Smithsonian Institution by the Conservation Analytical Laboratory (Van Dyke, 1980). Since the RH in a collection area is always higher than zero, these data present the minimum reduction in ethanol concentration for a given small loss of solution, again, assuming loss by simple evaporation. At 50% relative humidity the solution concentration lost by evaporation would be equivalent to about 95%v/v ethanol.

Equilibrium relative humidity (ERH) and water activity (Aw).—Another quantity that is directly related to the vapor pressure of the solution components is relative humidity and its near-identity, water activity. Relative humidity is found by dividing the vapor pressure of water exerted by the solution (P_{water}) by the vapor pressure of pure water at the same temperature. The solution can be said to have a relative humidity (RH), but RH is so commonly thought of as applying to a gas phase that the alternate designations of equilibrium relative humidity or water activity are more commonly employed. Although Aw is not identical to ERH, at room temperature and normal atmospheric pressures it deviates by no more than 0.2% (van den Berg and Bruin, 1981; Gál, 1972). Consequently, for all but the most exacting work they can be considered equivalent. ERH as a function of volume percent ethanol is tabulated in Table 3 and shown in Figure 6.

If the relative humidity in the air into which the solution is evaporating is higher than this equilibrium value, then water will condense into the solution rather than evaporate from it. For example, the data show that a 70%v/v solution will absorb moisture from the air if the ambient relative humidity is higher than 78.1%.

Osmotic pressure (Π).—Interestingly, the first published account of an osmotic phenomenon involved pressures developed across an animal membrane separating alcohol solutions. In 1748 Jean-Antoine Nollet demonstrated that when water and alcohol are separated by an animal bladder membrane, a pressure would develop

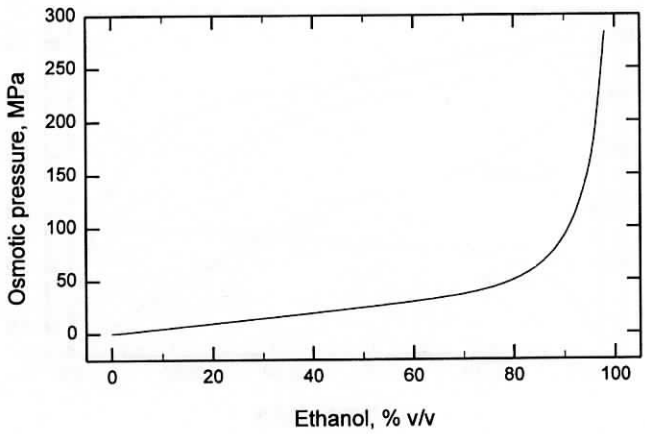

Figure 7. Theoretical maximum osmotic pressure as a function of volume percent concentration of the solution at 25°C.

due to movement of water into the alcohol (Glasstone, 1946). The requirement for stepping specimens through increasing concentrations has been noted by several authors (Fink, *et al.,* 1979; Laframboise, *et al.,* 1993) although this requirement has not always been linked with a need to limit osmotic pressures within specimens.

The maximum possible osmotic pressure due to water transfer through a semipermeable membrane as a function of ethanol concentration is shown in Figure 7. These values are calculated from the formula (Glasstone, 1946):

$$\Pi = \frac{RT \ln\left(\frac{p^0}{p}\right)}{\bar{V}}$$

where:

Π = osmotic pressure
R = universal gas constant
T = temperature, Kelvin
p^0 = vapor pressure of pure water
p = partial pressure of water vapor
$\bar{V}$ = partial molar volume of ethanol

This is the theoretical maximum which assumes that the barrier (e.g., a cell wall) is a perfect semipermeable membrane and that the internal fluid is pure water. In addition, possible influences of other dissolved components have not been considered. Nevertheless, it is clear that the osmotic pressure rises steadily with ethanol concentrations for solutions below about 75%v/v and begins to rise much more rapidly at concentrations above about 80%v/v. These facts suggest that, from considerations of osmotic pressure, solutions with approximately equal concentration increments are appropriate for stepping specimens up to higher ethanol concentrations, up to about 75%v/v. At much higher concentrations, progressively smaller concentration steps are indicated. The theoretical pressures given here are huge in comparison to those leading to shrinkage or swelling of marine

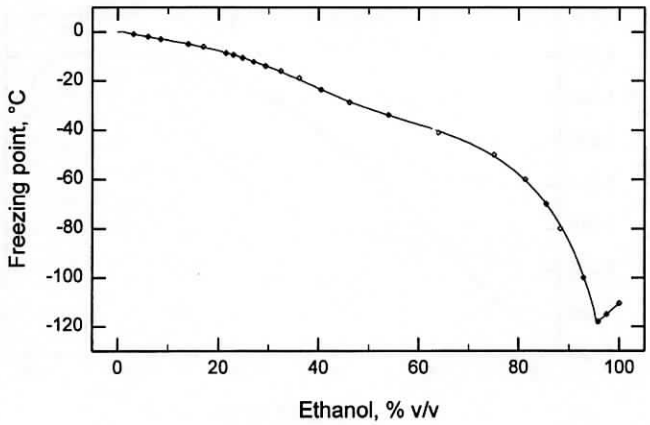

Figure 8. Freezing point as a function of volume percent concentration of the solution. Points are data from Mellan (1977).

zooplankton in fixation and preservation fluids (Steedman, 1976). The pressures Steedman quotes are on the order of just a few MPa (1 osmole = 2.3 MPa).

Freezing point and boiling point (Tf and Tb).—Freezing points and boiling points are shown in Figures 8 and 9 respectively. Freezing points were interpolated from data in Mellan (1977). Data for boiling points are from Rieder and Thompson (1949) as reported in Chu *et al.* (1956). Although these do not have direct applicability to conservators and collection managers, they are of potential interest in that they could be used as indicators of alcohol concentration, provided the concentration is known to be under 95%v/v. The freezing point may be especially useful since it can be determined using just microlitre quantities of sample. In addition, the effect of impurities will be different for the different physical parameters that could be used for indicating concentration. Thus, the difference in indications of concentration found when different properties are measured could provide some characterization of the impurities present.

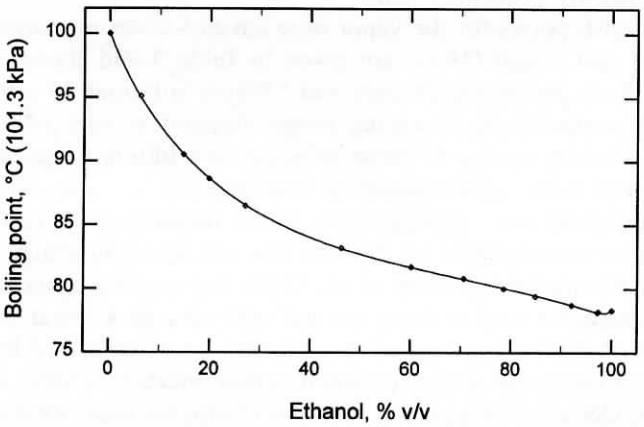

Figure 9. Boiling point at 101.3 KPa as a function of volume percent concentration of the solution. Points are data from Chu et al. (1956).

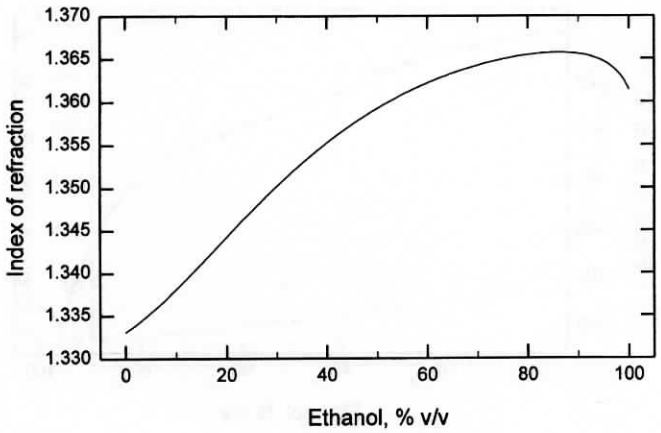

Figure 10. Refractive index as a function of volume percent concentration of the solution.

Refractive index (n).—The refractive index (Weast, 1976) is shown as a function of volume percent ethanol in Figure 10. It is clear that volume percent is not uniquely defined by many values of refractive index, which is the reason that refractive index is not useful, by itself, for indicating ethanol concentration. It is possible that, with adequate experimental work, refractive index could be used in conjunction with other properties indicating concentration to provide information on the nature of impurities present. Another point of interest is that, in the area of 80 to 90%v/v, refractive index is independent of concentration. This has implications when using the method of introducing a stream of solution of unknown concentration into a solution of known density to determine density differences. If both test and standard solutions are in the range of 80 to 90%v/v, it becomes difficult or impossible to see the test solution.

Flash point.—The flash point is the lowest temperature at atmospheric pressure at which fuel vapor is produced in sufficient concentration to support combustion once ignited (ASTM, 1988). "Closed cup" flash points are equivalent to the lower limit of flammability (Thorne, 1976).

Closed cup flash points for the vapor over ethanol-water mixtures based on the data in Nabert and Schön (1963) are given in Table 3 and shown in Figure 11. Note that the flash points for 95%v/v and 75%v/v solutions of ethanol in water are near room temperature. Lowering room temperature can reduce the risk of fire involved with the storage of these solutions in addition to generally reducing rates of evaporation and of deterioration reactions.

Dielectric constant (ϵ).—Interpretation of pH measurements taken in ethanol-water solutions is complicated by the fact that pH varies in a non-linear fashion with ethanol concentration (Waller, et al., 1993). For a given concentration of acid and ratio of conjugate acid to base, the pH will vary as a linear function of the inverse of the dielectric constant of the solution for solutions of less than about 60 to 70%v/v ethanol (R. Stairs, personal communication, 1996). Consequently, knowing the dielectric constant as a function of ethanol concentration is essential for interpreting concentrations of acid-base equilibrium pairs based on pH measurements from solutions of different ethanol concentrations. The dielectric con-

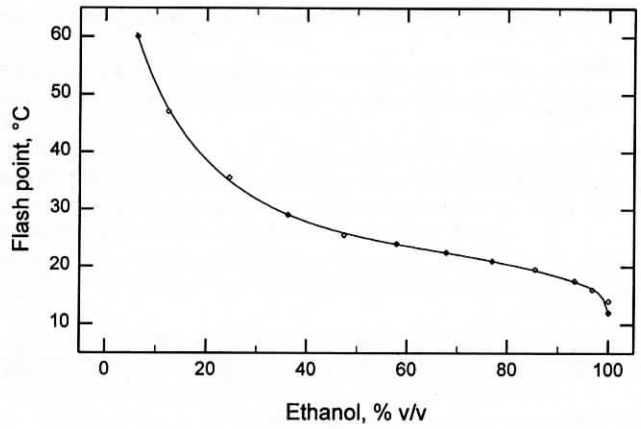

Figure 11. Closed cup flash point as a function of volume percent concentration of the solution. Points are data from Nabert and Schön (1963).

stant, interpolated from data in Åkerlöf (1932), is given in Table 3 and shown in Figure 12.

Antiseptic effect.—Although it is not strictly a physical-chemical property, the effect of ethanol concentration on biological activity is certainly a property of interest to collection managers and conservators. Data are not readily available on the effect of ethanol on all organisms of interest. Here, we have considered the bactericidal data available on ethanol as an indicator of antiseptic effect. Morton (1977) reviewed the times required for inhibition and killing of bacteria, fungi and viruses for various concentrations of ethanol and other alcohols. Information on non-sporing bacteria is most complete and was used to generate Figure 13. It is evident that ethanol solutions are most strongly antiseptic in the range of 50 to 80%v/v ethanol and that the time required to kill bacteria increased at both higher and lower concentrations. It is worth noting that these times are all very much shorter than the duration of specimen preservation that interests us.

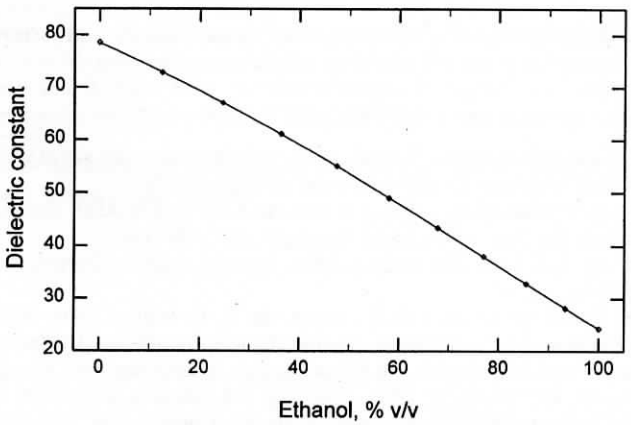

Figure 12. Dielectric constant as a function of volume percent concentration of the solution. Points are data from Åkerlöf (1932).

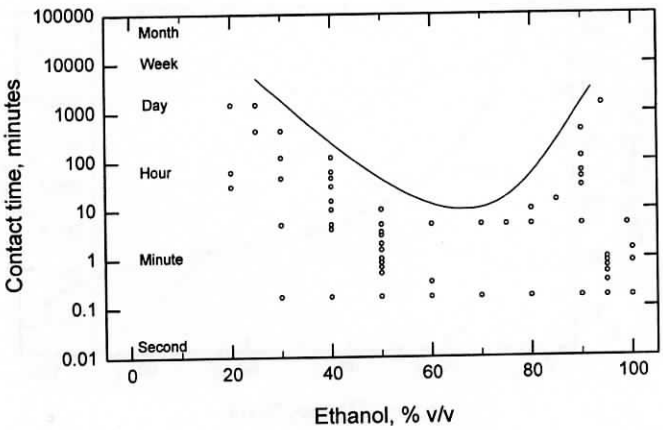

Figure 13. Bacterial mortality times as a function of volume percent concentration of the solution. Points are data from Morton (1977).

Conclusions

The relations between volume percent, weight percent and mole fraction as expressions of ethanol-water solution concentration have been given. Data on fifteen physical-chemical properties of ethanol-water solutions, in most cases over the entire range of 0 to 100% have been collected. Considerable data on other properties and characteristics of ethanol-water solutions are available. This includes, for example, information on solubilities of organic and inorganic compounds, of gases, such as oxygen, of the behavior of proteinacious materials in solutions, and so on. We have not included all of this information, either because the data are too fragmentary, are not useful when presented out of context of the original research, or are not thought to be of general interest. We hope that this paper will be a convenient source of information for anyone working with ethanol-water solutions as fluid preservatives, especially those who are contemplating or conducting research on their stability and efficacy.

Acknowledgments

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Thomson, G. 1986. *The Museum Environment,* 2nd ed. Butterworths, London, 293 pp.

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