1975

Stability, packaging, storage, and sterilization of a medicated dry foam

Armen Gostanian
University of the Pacific

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STABILITY, PACKAGING, STORAGE, AND STERILIZATION
OF A MEDICATED DRY FOAM

A Thesis
Presented to
the Faculty of the School of Pharmacy
The University of the Pacific

In Partial Fulfillment
of the Requirements for the Degree
Master of Science

by
Armen Gostanian, Jr.
July 1975
This thesis, written and submitted by

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Dated July 31, 1975
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A. G.

The University of the Pacific
Stockton, California
July 1, 1975
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CHAPTER I

INTRODUCTION

Denuded skin areas (e.g., partial and full thickness burns, severe traumatic injury, various skin diseases) present problems with respect to application and removal of conventional topical dosage forms. With these problems in mind, a highly water soluble, flexible film for topical application was developed (1-3). The film adheres to denuded and moist skin, dissolving quickly and releasing the active ingredient.

Both in vitro and in vivo studies were performed to evaluate the film. When comparing the film to commercially available topical antimicrobial products, in vitro and in vivo methods showed that the film was as effective as conventional bases for mafenide and gentamicin and more effective for silver sulfadiazine and silver nitrate. Also, preliminary stability and packaging studies indicated that the film, if properly packaged and stored, has marketing potential.

The basic unmedicated formula consists of dextran, sorbitol, Miranol 2 MCA Modified, and water. The major advantages of the film are 1) fast release rates, 2) ease of application to denuded skin without aggravation, and 3) lack of necessity for removal, hence no aggravation. However, it is extremely sensitive to moisture and elevated temperatures which is a major disadvantage. Excessive humidity will solubilize the film or make it too tacky to handle while very low humidity causes the film to lose its flexibility. Also, elevated temperatures remove moisture from the film and cause it to lose flexibility much as with
This study was designed to evaluate further this new dosage form with respect to:

1. Modification of formula to minimize moisture sensitivity.
2. Determination of a suitable packaging material.
3. Determination of proper storage conditions.
5. Determination of a suitable sterilizing method(s).

Percutaneous Absorption

Because there are many areas of controversy surrounding the concept of percutaneous absorption, it is literally impossible to make positive statements about all of its aspects. Nevertheless, a review of commonly accepted generalities does serve as a reference point for a discussion of topical dosage forms.

Percutaneous absorption, as defined by Rothman (4), Collaizzi (5), and Barr (6), is the penetration of substances from the outside into the skin and through the skin into the bloodstream. However, Blank (6,7) states that an actual definition of percutaneous absorption is not necessary, but rather it is more important to know how drugs move into and through the skin. It is here that so many areas of controversy arise, i.e., routes of entry, factors affecting the rate of absorption, factors affecting the extent of absorption, and methods of evaluation.

Historically, there are three periods and, hence, three schools of thought regarding the mechanisms of percutaneous absorption (6). These are 1) the period up to 1877 in which it was accepted that gases and volatile substances penetrated the skin easily; 2) 1877-1900 in which...
Fleischer's theory that the skin is impermeable to all substances predominated; and 3) 1900 to the present in which Fleisher's theory has been disproved and in which much research has been done to determine the mechanism of and the factors affecting percutaneous absorption. This last period will be reviewed here.

**Routes of Entry**

Bliss (8) and Mali (9) claim transepidermal entry through the *stratum corneum* to be the major route because its surface area is 100 to 1000 times that of the transappendageal route. Shelmire (10) and Gemmell and Morrison (11), on the other hand, claim that the major route is via the skin appendages (pilosebaceous regions and sweat glands). They base this on the observation that hair follicles possess preferential staining properties, perifollicular wheals develop, and dyes diffuse rapidly through the sweat glands. Most investigators (4-6, 12, 13) agree that both processes are involved and that some drugs may have a greater affinity for one route over the other. Furthermore, Scheuplin (12) and Arita *et al.* (14) state that for drugs capable of penetrating the skin, there are two stages. In the initial or rapid stage, the transappendageal route is the primary method of entry and in the second or steady state stage, the transepidermal route is the major method. All investigators agree that before a route of entry comes into play, the drug must first be released from the vehicle, transferred to the skin, and then have the ability to penetrate the skin.

**Factors Affecting Absorption**

When developing a topical dosage form, the investigator is essentially working with three separate entities: the skin, the drug,
and the vehicle. Separately, each possesses certain properties which affect percutaneous absorption. However, in actuality, all three entities necessarily comprises a skin-drug-vehicle system such that the final effect is a composite of all the separate factors working together. Some of these factors such as hydration of skin, drug form, vehicle composition, and concentration of drug in the vehicle can be altered; others, namely those involving the anatomy of the skin per se, cannot be changed. The purpose of the skin is primarily that of protection and this protective barrier must be overcome to effect percutaneous absorption.

Skin factors affecting percutaneous absorption are related to the structure and physical/chemical properties of the skin components. While these properties will vary in different sites and from person to person, some generalities seem to hold. The major barrier to absorption is the s. corneum with its layer of lipids (4,8,15-18). Some feel that the major barrier is really between the s. corneum and the dermis (6,10,14). However, all agree that the major barrier is electronegative in nature and consists of keratin and other lipid materials. Interspersed throughout the s. corneum are the various appendageal ducts which are filled with sebaceous or lipid substances. Thus, regardless of the route of entry, a drug must be partly soluble in lipids in order to penetrate. Likewise, undissociated drugs will penetrate well since negatively charged drugs will be repelled and positively charged drugs attracted and bound to the charged barrier (4,6,11,14). Rothman (4), however states that di- and trivalent ions will penetrate to some extent because they are partly lipid soluble. Keratin, a sulphydryl protein which adsorbs water and other polar compounds, can potentially interact
to bind with various drugs further complicating the predictability of percutaneous absorption for any drug (6).

Other skin factors affecting absorption relate to difference in sites and the general condition of the skin. For example, the forehead, presternal area, and the back demonstrate the greatest permeability. Thicker areas such as the soles of the feet and palms of the hands are the least permeable (8,10,19-21). Hydration from moisture and sweat will increase absorption because the keratin swells and becomes more permeable and the intercellular pores become larger (4,6,8,10,13,17,20, 22-28). Aged skin, due to atrophy and dehydration, is less permeable (6,8,19,26) while young skin and skin of women show increased absorption, presumably because of greater hydration and thinness (8,19). These statements are general and individual variation must be considered.

All of the above factors relating to the skin do not affect percutaneous absorption if the **stratum corneum** is removed purposely or by disease or traumatic insult. In such instances, all drugs penetrate easily and readily and this fact substantiates that the **stratum corneum** (or zone beneath it) is the primary barrier to absorption (4,6,10,19,20,26, 29).

The skin factors can be altered by removal of the skin, hydration, and emulsification. Removal, of course, is not desirable except in experimental conditions. Thus, it is the hydration and emulsification aspects that product developers try to attain for topical preparations. Once a drug has crossed the major barrier, the lower layers of the epidermis and dermis offer little resistance to absorption into the bloodstream and subsequent pharmacological affect. The factors which now enter the picture are protein binding, receptor specificity, rate of
metabolic inactivation, and rate of excretion (30).

**Evaluation of Drug Product**

Two fundamental processes must occur before a topical drug can be effective. First, the drug must be released in therapeutic concentrations from the vehicle at an adequate rate and then the drug must be capable of penetrating the skin. Thus, the product developer must employ methods to evaluate the rate and extent of release and subsequent absorption. Wurster and Seitz (31) have stated that vehicles which are porous, i.e., have a high surface area to weight ratio, will have faster dissolution rates and hence, faster release rates. In addition, Marcus *et al.* (32) showed that the release of an active ingredient is not influenced by but is independent of the presence of other noninteracting drugs, since the thermodynamic activity of the drug in a vehicle is the only significant factor governing rate of release and absorption. Thus, they concluded that incorporation of two or more noninteracting drugs into a vehicle will allow decreased doses and side effects resulting in a less expensive product.

Topical products can be evaluated by *in vitro* and *in vivo* methods (6,7,11,21,26,27,33,34). *In vitro* methods include 1) diffusion without a membrane using chemical-physical and microbiological methods; 2) diffusion through a membrane using chemical-physical and microbiological methods; and 3) tracer methods using radioactive substances, dyes, and fluorescent compounds. These methods are mainly used to evaluate release rates. Because the continuous phase in the diffusional media is aqueous, direct correlation with *in vivo* results is not possible. Nevertheless, they do provide an excellent guide for determining the relative effectiveness of a vehicle (4,6,11,21,26,27,33-37).
Diffusion methods without a membrane usually employ an agar gel in a tube or plate. For chemical-physical methods, the agar media may contain a reagent that produces a color when in contact with the drug (e.g., salicylic acid with ferric chloride). The rate and extent of release is calculated based on the diameter of the color zones in millimeters plotted against time. This method is simple and inexpensive, is not time consuming, and does not require precautions such as aseptic technique or protection from radiation. However, it is not a good quantitative method and only drugs which will produce color reactions can be used (7,11,21,26,33-36).

Microbiological diffusion employs nutrient agar medium which is seeded with a specific test microorganism. Release of antimicrobial drugs can then be evaluated by measuring the zones of inhibition. The diameter of the zones in millimeters is a measure of the release rate. This method provides a good procedure to evaluate release of antimicrobials (7,11,21,26,33-36,38-40). Shelmire (10), Billups and Patel (21), Gemmell and Morrison (11), Zheutlin and Fox (41), and Scheuplin (12) all state that a topical antimicrobial drug does not need to penetrate the skin; hence, if a microbiological procedure indicates that the drug is released sufficiently to inhibit microbial growth, this method of evaluation is very useful for these agents. However, they do recognize the fact that the skin, because of its different composition from an aqueous agar gel, may alter the extent of release such that therapeutic concentrations may not reach the site of action in vivo.

Diffusion methods using a semipermeable membrane such as a dialysis cell or agar tube covered with a membrane provide two distinct advantages over nonmembrane procedures. These methods 1) resemble a
more true-to-life situation in which the skin is the membrane and 2) allow quantitative analysis of the receptor chamber using various chemical methods. For chemical-physical methods, many types of diffusion models using a membrane have been devised. Some employ very elaborate and sophisticated instrumentation while others are simple and straightforward (41-53). Membranes made of sheep bladder or isolated skin have been claimed to resemble true skin better; however, one must realize that there is a great difference between dead and living epidermis (6).

The basic apparatus uses two chambers separated by a membrane. One chamber is filled with the receptor medium (usually aqueous) while the other contains an accurately measured quantity of the drug product. At various time intervals, exact quantities from the receptor medium are removed for analysis by any satisfactory method (21,45). The disadvantages of this method are that nondialyzable substances cannot be evaluated and if the product chamber contains a liquid medium, drugs in which the vehicle is highly soluble cannot be used (43).

For microbiological studies, an agar tube seeded with the microorganism is usually covered with a membrane and the product placed on top. Zones of inhibition are then measured to evaluate release (42). The advantage this procedure has over the nonmembrane method is that it resembles the true situation a little more closely. Again, quantification is not very good.

Of the three tracer methods, the use of radioactively labelled compounds is best. While dyes and fluorescent compounds are easily analyzed, the dye or fluorescent compound may not remain with the drug or they may alter the physical-chemical properties of the drug (6). However, labelling of a drug with a radioactive element can provide
evaluation of almost all therapeutic agents; trained personnel is the only major limitation. Nevertheless, the use of radioactive labelling is by far the best method to quantitatively evaluate the release and absorption of drugs (6,7,11,21,26,27,33-36).

The indiscriminate use of in vitro physical models and subsequent calculations can lead to misleading and erroneous results with respect to rate and extent of release (54). Chowhan and Poulsen (54), in their critical review, emphasize that the mathematical formulas to calculate release rates differ for solutions, suspensions, and semisolids. Furthermore, one must take into consideration the number of therapeutic agents present, changes in vehicle composition after application, and other parameters when devising an in vitro model.

For obvious reasons, in vivo methods should be used to evaluate a drug product, especially with respect to absorption, metabolism, and excretion (6,7,11,26,27,33). Methods used include measuring of a physiological response such as vasoconstriction, analysis of body fluids such as urine, loss of drug from the skin surface, histological procedures, and tracer methods. While in vivo methods are preferable, they do possess certain disadvantages. If lower animals are used, direct correlation with humans cannot be made; if humans are used, expense, cooperation, control factors, and health hazards become problems.

Physiological responses rely greatly on subjective evaluation (6,7,33) although McKenzie and Stoughton (55), McKenzie (56), and Vickers (57) developed acceptable vasoconstriction tests for corticosteroids. Washitaki et al. (18,29) used the vasoconstriction test for steroids and a vasodilatation test for carbinoxamine and obtained acceptable results for absorption. Rate of excretion, storage tendencies,
rate of entry into and out of the bloodstream, and rate of metabolism must be known for tissue analysis methods; otherwise, one must estimate time intervals for analysis (6,7,33). Using loss of drug from the skin is probably the worst in vivo method of all (6,7,33). In this method, the drug product is removed from the skin and analyzed for drug concentration. The difference at the time of application and at the time of removal is considered to be absorbed. However, the pitfalls in such a procedure are that it is difficult to remove the entire product completely, the drug may have complexed with skin components and not been absorbed, and the quantities one is working with are so small that errors in analysis are likely. Histological studies are not very valuable from a quantitative viewpoint (6,7,33). Disadvantages of dyes and fluorescent tracers were mentioned earlier along with some limitations of radioactive tracers. Other disadvantages of radioactive methods are 1) autoradiography indicates if a drug has reached a particular physiological site and now how the drug reached that site; and 2) urinary analysis only indicates that the drug is excreted in the urine and not the pathway into the urine. Thus, while in vivo methods are superior to in vitro methods, they are nevertheless only estimations and one must take this and specific disadvantages into consideration (6,7,).

Stability

Whittet (58) and Schou (59) have defined stability of a drug product as the period of time from completion of a preparation until it no longer fulfills specifications of the pharmacopeia or until a 10% loss in potency has occurred. Thus, stability of a drug product is an integral part of quality control. A stable product implies that at
least 90% of the labelled quantity of an active ingredient is in the product; that each dose will be uniform; that the product will maintain its appearance; that the drug will be made bioavailable; that the product is safe when used properly; and that the package and recommended storage conditions will not affect the product (37,60). A shelf-life of one year is considered fair while five years is considered the maximum since no finished product is entirely stable (60-62). Consequently, compendial regulations and guidelines for good manufacturing principles must be followed (59,63).

Instability or degradation is caused by chemical, biochemical, and physical factors (58,59,61,64-66). Whittet (58) and Smith (64) reviewed these degradation processes and methods to minimize or prevent them. Chemical and biochemical factors include oxidation, reduction, and hydrolysis reactions, racemization, specific biochemical reactions, and drug-package interactions.

Oxidation is defined as the addition of electronegative atoms or radicals or the loss of electropositive atoms or radicals. Addition of oxygen or removal of hydrogen are usually involved. Air and oxygen will cause oxidation of drugs such as oils, fats, phenols, and vitamins. Antioxidants will minimize oxidation as will removal of all air or oxygen from the package. The air or oxygen is usually replaced with nitrogen or carbon dioxide, although one must be aware of pH changes caused by carbon dioxide. A 10° increase in temperature increases oxidation rates two to threefold; thus, temperature control during storage is important. Absorption of radiation such as ultraviolet or gamma rays also increases oxidation rates. Radiation sensitive drug products should therefore be stored in light-resistant glass containers or laminated aluminum foil.
Metal ions are catalysts in oxidation reactions and these should be complexed with EDTA or the like. Moisture and pH also facilitate oxidation. Thus, adjustment of pH and use of moisture impermeable containers such as aluminum laminates may be required to reduce or eliminate oxidation reactions.

Reduction reactions are defined as the addition of electropositive atoms or radicals or the removal of electronegative atoms or radicals. Again, oxygen and hydrogen are most often involved. These are not as common as oxidation reactions and usually involve the reduction by light of metal ions such as gold, silver, and mercury. Use of light-resistant containers will usually prevent these degradative reactions.

Racemization may result in loss of potency and it obviously involves those drugs which have only one therapeutically active enantiomer. Racemization requires moisture and thus, moisture impermeable containers aid in minimizing or preventing racemization reactions from occurring.

Degradative biochemical reactions can be caused by enzymes or microorganisms. Removing moisture, where possible, will prevent enzymatic degradation. Storage at low temperatures, use of preservatives, and protection from moisture will usually eliminate microbial growth and subsequent degradation caused by microorganisms.

Drug-package interactions such as sorption and leaching are usually associated with plastic containers. Selection of a noninteracting plastic or a different type of container will help prevent or minimize these reactions.

Physical factors usually cause degradation indirectly by eliciting one or more of the above described chemical reactions. However, physical
factors may also directly affect drug products in a number of ways. Moisture will solubilize or cause caking of hygroscopic, effervescent, and deliquescent products while changes in humidity may cause these products to gain or lose significant amounts of moisture. Increased storage temperatures might cause melting of certain drug products while decreased temperatures lead to precipitation of biologicals or freezing of certain dosage forms. Further, high temperatures many times employed during manufacturing and sterilizing methods may degrade or even char certain drugs and packages such as plastics; similarly, radiation sterilization destroys drugs such as atropine sulfate, heparin, and progesterone as well as packages consisting of polyethylene and soft glass. Breakage, cracks, corrosion, and sealing defects of packaging materials cause obvious instability since the drug product is no longer protected from the environment. Finally, formulas themselves may cause cracking, creaming, caking, drying, and aggregation resulting in an unstable product. In order to assure stability, these physical factors must be eliminated or avoided by defining proper storage conditions, employing appropriate manufacturing and sterilizing procedures, selecting suitable packaging materials, and using stable formulations.

While one can theoretically minimize or prevent instability mechanisms, the product developer must nevertheless perform stability tests to prove that the product is stable. The tests must be performed on the final packaged product in order to be valid (58,59). The most common experimental procedure involves storage at various temperatures with or without varying relative humidities; the product is then analyzed by various chemical or microbiological procedures as well as physical inspection (37,39,40,61,67-76). The method chosen for analysis is
somewhat dependent on the nature of the drug. Chemical methods such as chromatography and photometry are the most common; colorimetry with photometry are used for drugs which form color reactions with a reagent. Agar diffusion with subsequent measurement of zones of inhibition is most common for antimicrobial products (39,40,59,61,67).

The rate of degradation for most drug products follows first order kinetics (66,71). That is, the concentration of the drug decreases with respect to time; the rate at which it decreases is dependent on the concentration. The basic equation for a first order reaction is

$$\frac{dC}{dt} = kc$$

where C is the concentration at time t and k is the first order velocity constant. In order to calculate k, the equation has to be changed via integration to a workable form as follows:

$$\int_{C_0}^{C} \frac{dC}{C} = -k \int_{0}^{t} dt$$

(Eq. 1)

$$\ln C - \ln C_0 = -k(t-0)$$

(Eq. 2)

or

$$\ln C = \ln C_0 - kt$$

(Eq. 3)

or

$$\log C = \log C_0 - \frac{kt}{2.303}$$

(Eq. 4)

or

$$k = \frac{2.303}{t} \log \frac{C_0}{C}$$

(Eq. 5)

where $C_0$ is initial concentration, $C$ is the concentration at time $t$, $0$ is initial time, i.e., time zero, and $k$ is the specific degradation
constant (66,71). Once \( k \) has been calculated, the concentration at any time \( t \) can be calculated. The shelf-life of a drug product can then be predicted from performing stability tests for relatively short periods of time, assuming that the product is still in a usable form and that concentration is a measure of activity.

Temperature affects degradation rates as shown by the Arrhenius equation below (59,66,70,71,76,77).

\[
\begin{align*}
  k &= \frac{A e^{-E_a/RT}}{2.303} \quad \text{(Eq. 6)} \\
  \log k &= \log A - \frac{E_a}{2.303RT} \\
  \text{where} \quad k &= \text{specific rate of degradation} \quad -1 \\
  R &= \text{gas constant (1.987 calories degree moles}^{-1} \text{moles}^{-1} \\
  A &= \text{entropy constant} \\
  E_a &= \text{energy of activation} \\
  T &= \text{absolute temperature}
\end{align*}
\]

This is the reason that most stability studies are carried out at various elevated temperatures and then the results are extrapolated to room temperature. This method allows one to use relatively short storage periods of months as opposed to uneconomical and impractical storage test periods of one year or more. Results of elevated temperature studies are valid when the degradation is due only to thermal phenomena and not due to photochemical, microbiological, freezing, or other physical factors. For these latter instances, long term storage studies and the use of appropriate order equations are required (66).
Packaging

The function of a package is to preserve goods so that they get to the consumer for their original intended purpose (65,78). In addition to providing mechanical and environmental protection, packaging materials must also be functional, inert, and available at a reasonable cost. Other desirable features include lightness in weight; ease of storage, use, and labelling; and aesthetic appearance. The packaging material should also be nonflammable, noncorrodable, nontoxic, odorless, and be ecologically advantageous. Availability in various sizes, shapes, and colors further adds utility to a packaging material.

The common types of materials used to package pharmaceuticals are glass, paper, plastics, and metals including aluminum foil. The manufacturer must select the most appropriate package for each type of pharmaceutical; all packages possess some disadvantages. While glass meets the main criteria, its major disadvantages are that it is easily broken, not as versatile as plastics and aluminum foil, and it is fairly heavy and bulky. Paper packages are primarily used for bandages, gauze, or other types of products which are not affected by atmospheric constituents.

Plastics provide the most versatile type of packaging materials as they possess most of the suitable properties mentioned above (65,79-84). The major disadvantages of plastics are permeation, leaching, sorption, chemical reactivity, and alteration of structure (79-82, 84-93). Permeation refers to the passage of gases, microorganisms, drug constituents, and air constituents through the package. Obviously, permeation can occur in either direction. Similarity of chemical structure between the plastic and the permeant increases permeability as do
elevated temperatures and increased atmospheric vapor pressure. A decrease in plastic crystallinity will also increase permeation. Increasing the thickness of the plastic does not necessarily decrease permeation. Effects of permeation are an increase in chemical degradation rates, pH changes (due to carbon dioxide), changes in drug potency, and biochemical degradation due to microbial contamination. Leaching is defined as the migration of an additive from the plastic into the drug product. While pure plastics can exhibit leaching of components, the compounded plastics create the major problems. Stabilizers are the most commonly leached substances and cause most of the toxic reactions due to plastics. The pH of the drug product must be compatible with the plastic to minimize leaching.

Sorption is defined as the migration of a component of a drug product or the drug itself into or onto the plastic. It is a physical/chemical process and usually involves amorphous zones. Consequently, polar plastics exhibit greater sorptive properties than nonpolar plastics. Low molecular weight and ionic or electrically charged drugs have greater sorptive tendencies while steric and internally bonded drugs are sorbed less. A decrease in pH increases sorption of acidic drugs. If the plastic and solvent system are both polar or both nonpolar, increased sorption occurs because like systems cause swelling and open up sorptive passages. Increased drug concentrations and increased temperatures also facilitate sorption. It is the chemical structure of the drug product and plastic which determines if sorption will occur; all other factors only affect the magnitude of sorption. If both adsorption and absorption occur, the quantity adsorbed is insignificant with respect to the quantity absorbed. Some documented examples of
drug-plastic sorption are those of nylon with some antimicrobials; polyvinylchloride will sorb benzalkonium chloride while polyethylene will sorb some steroids. Low density polyethylenes have been noted to sorb scopolamine, pilocarpine, ouabain, and other similar compounds (85).

Chemical reactivity between drug and plastic may result in physical changes of the plastic such as discoloration or deterioration. For example, benzyl alcohol, benzaldehyde, and paraldehyde will etch and dissolve polystyrene; certain oils will dissolve polyethylene; fluorinated hydrocarbons attack polyethylene, polystyrene, and polyvinylchloride. The choice of the right plastic polymer will help avoid such problems (85,87,89). Alteration is usually due to these four drug-plastic interactions. For example, gases or vapors may cause swelling or collapsing of the package depending on which way the gas permeates. Heat, UV light, ethylene oxide, and other sterilization procedures may also deteriorate some plastics (85,87,89). Provided no contraindications with respect to these disadvantages are present, Estevez et al. (81) state that plastics should be used when:

1. Plastic offers particular functional advantages.
2. The drug product is so expensive that breakage must not occur.
3. The drug product is so dangerous or obnoxious that breakage must not occur.
4. The risk of breakage is high and the consequences of broken glass are very dangerous (e.g., in nurseries, etc.).
5. Mailing and transportation costs are based on weight and thus, are high.
6. Use of printed containers is advantageous.
7. Specific shapes are desired.
8. Plastic is the only practical choice.
Drug manufacturers must ascertain the potential of drug-plastic interactions as well as the stability aspects of the plastic itself (81). While many authors have cited some specific drug-plastic interactions (62,79,80,85,88-92,94-96), any time a new drug-plastic system is used, storage of the packaged drugs at various conditions and subsequent analysis must be done (62,87). Stability of drug products has already been discussed.

To determine changes in the plastic, impact resistance, average tensile strength, average ultimate elongation, melt index, infra-red analysis, and moisture permeability tests must be performed before as well as after storage. In addition to these tests, physical inspection of the package with respect to rupture and/or swelling must be done to determine stability.

Toxicity due to plastics appears to have been overestimated (81, 86,90). However, toxicity can occur directly from actual contact with the plastic as well as indirectly via contents of the package. The most common acute toxic reaction caused by pure plastics is sensitivity; however, it is rare. Compounded plastics as used in catheters or other internal devices, can cause thrombosis, embolism, thrombophlebitis, and necrosis. Also, if the plastic is contaminated, septicemia and bacteriuria may occur. In addition, such devices have been known to break-off in the body and cause physical damage to the circulatory system, urinary tract, etc. Direct toxicity is usually due to stabilizers present in the plastics. Chronic toxicities are manifested chiefly by neoplasms. While not yet seen in man, various malignancies in other animals have been caused by plastics such as polyethylene, polyvinylchloride, nylon, and others. Various mechanisms have been suggested as to the
carcinogenicity of plastics (90). Indirect effects are usually due to plastics used as occlusive dressings where excess percutaneous absorption may occur. Also, solutions and/or blood stored in plastics may leach some of the harmful additives (90).

Metals are used mainly for packaging aerosols and nuclearpharmaceuticals. In addition, laminated aluminum foil, because of its impermeability and light weight, is the package of choice for moisture sensitive, light-weight products (65,78,83,97). It must be sterilized via gamma-radiation (83,84,98,99). Laminates, such as lacquers, vinylchloride, and polyethylene, are used to provide a heat seal. Because polyethylene adds mechanical support, it is the most widely used (100). The seal should be peelable so that sterile products can be removed aseptically. Hughes (100) adds that the heat seal is the most common cause of defective aluminum foil packages. The sealing process must be tested by immersion in water for five minutes and then inspecting the package visually for moisture. Hughes also states that laminated aluminum is the most moisture impermeable flexible package. To assure impermeability, at least 0.018 mm and preferably 0.03 mm gauge foil should be used (83,100). Often the outside of the foil is coated with cellophane to increase impermeability and add an aesthetic sheen to the package (101). In general, laminated aluminum foil meets almost every suitable property cited.

**Sterilization**

Sterilization is a process by which all microorganisms are killed or removed such that they are no longer detectable in standard culture media in which they are known to grow (102,103). Bowman (49) adds that
sterilization does not necessarily kill all microorganisms but rather reduces them in number such that they cannot multiply in favorable media and thus, the sterilized article is made safe for use. The common sources of contamination are raw materials, equipment, containers, personnel, environment, and the user (102).

There are numerous methods of sterilization. Boucher (108) defines disinfection as a method which destroys disease causing microorganisms; only when it becomes powerful enough to kill all types of microorganisms, is it a sterilizing method. The AOAC test established by the USDA is used to determine if a disinfectant is, in fact, a sterilant.

The various sterilizing methods possess specific advantages and disadvantages with respect to usefulness. The properties of the articles to be sterilized and the properties of the sterilant will determine what method is best (102). Some generalities apply to all sterilization methods. These include testing, certification, and adequate record keeping of sterilization cycles. In addition, quality control must be employed using suitable indicators and testing media as well as strict adherence to procedural and operational instructions. The proper training of personnel is imperative (103,105,109,110).

Autoclaving is an easy and effective sterilization method without toxicity or residues. It is easily controlled and measured and is economical after initial installment. A large majority of articles can be sterilized with this method. However, oils, greases, powders, and similar substances cannot be penetrated by steam and sharp instruments are dulled. All air must be removed from the chamber and some drugs are deteriorated by high temperatures and steam. The mechanism of action is
enzyme inactivation and protein denaturation. In general, autoclaving is the best method of sterilization when it can be used (103,108).

A dry heat oven with air circulation becomes a very useful method of sterilization. Temperatures of 160° - 170° for 1-3 hours will kill most microorganisms and destroy pyrogens (103). Again, enzyme inactivation and protein denaturation are the mechanisms of action. The major advantages of dry heat are that instruments are not blunted or corroded, and oils, powders, and steam sensitive drugs can be sterilized. Its disadvantages are long exposure times and high temperatures (103).

While not practical for hospitals or laboratory use, irradiation with X- or gamma rays is a very effective means of sterilization (98,99, 103,108,111). Sterilization occurs directly via ionization and indirectly via diffusion of radicals (108,111). Gram negative bacteria are very sensitive to radiation while viruses, yeasts, fungi, and spores exhibit varying degrees of resistance. Major sources of these sterilizing rays are Co-60 and Cs-137 (99,108). The advantages of irradiation sterilization include quality control as to number and energy of the radiations; the products do not become radioactive; it is effective against most microorganisms; and it penetrates most packaging materials. Its disadvantages are that trained personnel are required and some packages are destroyed. Factors affecting radiation sterilization are presence of oxygen or other protective compounds, physical state and water content of the microorganisms, and temperature (108). B. pumilus and B. stearothermophilus are the recommended biological indicators (104,108,109,111). Radiation is the best method to sterilize topical drug products, especially enzyme preparations, and is the best method to sterilize laminated aluminum foil packages as mentioned earlier. Dietz
(99) has reviewed the aspects of contract sterilization and discussed the many advantages it possesses, including relative inexpensiveness. It is best to sterilize the finished, packaged product (98,99).

Ultraviolet radiation photons of high energy kill microorganisms via DNA inhibition (112). Maximum bactericidal effects occur at 240 to 280 nm; 253.7 nm is used commercially because most UV lamps emit 95% of radiation in this latter wavelength (108,112). Applications are limited in that UV rays have poor penetrating power and thus, can only be used on clean surfaces with simple geometry and at short distances (108). Also, certain plastics polymerize on exposure to UV rays (108,112). Its major use is in the sterilization of room air for which it is highly effective (113-116).

Filtration employs the use of screen filters which have a pore size of 0.22 microns or less (117). These filters are made of cellulose, synthetic polymers, or silver films. The pore volume constitutes approximately 70-85% of the filter diameter so that high flow rates are achieved (118). The filters are inert, maintain their original integrity, do not impart fibers or debris, possess minimum adsorption, and are resistant to water, dilute acids, and hydrocarbon solvents. They do not affect the pH of filtered solutions and are very useful for sterilizing drug products which are sensitive to heat or cannot be sterilized by other methods. Disadvantages are that the filters are not resistant to corrosives such as nitric acid, sulfuric acid, sulfur, and cyanide solution. Silver membranes undergo the usual silver reduction reactions (118).

Ethylene oxide (EtO) gas was first used as sterilizing agent in 1933 (119). Because of its high solubility, it penetrates most porous
materials (103,108,120-122). When conditions are appropriate, EtO kills all microorganisms via alkylation of sulfhydryl, amino, hydroxyl, carbonyl, and phenolic groups present in the organism (108,121,123-127). This alkylation, shown below, causes irreversible metabolic changes in the organism.

\[
\text{EtO} \quad \text{viable organism} \quad \text{killed organism}
\]

Dessicated occluded spores provide protection from EtO's lethal effects (112,124); however, if articles are cleansed of debris, usually the occlusivity is interrupted and the organism becomes susceptible (125). Sykes (112) stated that the two most resistant bacteria to ethylene oxide are Staphylococcus and B. anthrroides and yet, at a concentration of 500 - 750 mg/L of EtO for 6-7 hours, these organisms were killed. Because EtO is explosive, it is usually diluted with carbon dioxide, nitrogen, or Freon 11 or 12 (103,108,112,122,128). These diluted mixtures require longer exposure times than pure EtO (128). The conditions of an EtO sterilization cycle vary with dilutions. While different conditions have been used successfully, all investigators state that certain procedures will afford greatest efficiency (103,105,108,112,120-124,128,129). Elevated temperatures, i.e., 50°, relative humidity of 30-80%, exposure times of 4-12 hours, and EtO concentrations of 450 mg/L to 1000 mg/L are recommended. In addition, an aeration time of 100 hours at room temperature or 8-12 hours at 50° should be used. Further, vacuum for undiluted EtO and positive pressure for diluted EtO are recommended.
The main advantages of EtO are that it is effective against all microorganisms; the procedure is simple and causes little damage to articles. Further, it is useful for products sensitive to excess heat or steam; it penetrates most packages and is relatively inexpensive. Its disadvantages include explosiveness, long exposure and aeration times as well as toxicity. Direct toxic effects of EtO, its residues, and its two commonest derivatives, ethylene glycol (EG) and chlorohydrin (EC) have received much attention and some disagreement occurs among researchers (103,108,112,120,127,130-137). The general chemical reactions forming these derivatives are shown below (108,127).

\[
\begin{align*}
H_2C\text{CH}_2 + H_2O & \rightarrow H_2C\text{CH}_2 \text{HO} \text{ OH} \\
\text{EtO} & \quad \text{Water} & \quad \text{EG}
\end{align*}
\]

\[
\begin{align*}
H_2C\text{CH}_2 + \text{HCl} & \rightarrow H_2C\text{CH}_2 \text{HO} \text{ Cl} \\
\text{EtO} & \quad \text{EC}
\end{align*}
\]

These derivatives, like EtO itself, will dissipate from the article when allowed to aerate. Aeration time is dependent on the type of package being sterilized (103,108,112,120-122,128,138). The greater the solubility of EtO in the package, the faster the sterilization, but also the longer the aeration time required (120-122,134). To determine residual levels of EtO and its derivatives, many methods have been used (119,132,
Kaye (139) emphasizes that because of these toxic residues, overtreatment for "safety margin" purposes must not be used with EtO sterilization.

Boucher (108) has discussed various miscellaneous methods of sterilization. Included in his discussion are the use of B-propiolactone, alkylated glutaraldehyde, acid glutaraldehyde, iodine, formaldehyde and others. All have specific applications but because of various disadvantages, they are not commonly used to sterilize pharmaceuticals.

Once a method of sterilization has been chosen, both indicators and sterility testing must be employed. Bruch (107) has stated that two basic types of indicators are available, biological and chemical/physical. He claims that biological indicators are the only indicators which can evaluate or integrate all sterilization parameters and that they provide the best assurance of sterility. Commonly used organisms as biological indicators are B. subtilis var. niger for wet and dry heat (104); B. pumilus and B. stearothermophilus for gamma irradiation (104, 108, 109); and B. stearothermophilus for EtO (104, 105, 109, 140).

B. stearothermophilus is one of the commonest biological indicators available commercially (109, 140). Brewer and Arnsberger (140) have developed a method which combines a chemical indicator and the biological indicator, B. stearothermophilus. The chemical indicator, 4-(4-nitrobenzyl) pyridine, forms a blue color when exposed to EtO. Thus, 4-(4-nitrobenzyl) pyridine will indicate if EtO has permeated the package and B. stearothermophilus will indicate if the concentration was sufficient to sterilize the article. The chemical reaction for 4-(4-nitrobenzyl) pyridine and EtO is shown below.
Sterility testing involves culturing samples of each lot in suitable media. Either direct inoculation of the sample into the media or solubilizing the product, filtering it, and then culturing the filter may be used (102, 104). The main purpose of testing is to create optimum conditions for microbial growth; thus, suitable media must be employed. The USP XVII (141) recommends Fluid Thioglycollate Medium (FTM) and Sabouraud's Media. However, Soybean Casein Digest (SCD) has replaced Sabouraud's Media in the USP XVIII (106). Sterility testing is not an absolute guarantee since cultivation of all viable microorganisms is impossible, sampling is many times inadequate, and failures in technique may occur (104). If a product contains an antimicrobial or preservative, it must be inactivated prior to testing (142). Abdou (143) has compared the appropriateness of seven test media. His results showed that peptone liver digest broth, peptone liver digest agar, and dithionite-thioglycollate media were superior with respect to cultivating all types of microorganisms. If more concurring data is accumulated, these media may well replace the more common ones. All regulations regarding sterility testing allow for accidental contamination (104).
When performing sterility tests and using biological indicators, both positive and negative controls should be used (142) and one must always recognize that none of these methods are an absolute guarantee that the entire load is sterilized (102, 105, 107, 110, 121, 124, 128).

Review of Objectives

A water soluble, flexible dry foam was developed for application to denuded skin areas (1-3). The foam demonstrated therapeutic equivalence when compared to commonly used ointment bases. However, the foam was physically unstable as deterioration and/or loss of flexibility occurred after storage at various conditions.

This study will improve the stability of the dry foam by modifying the original formula, determining a suitable packaging material, and defining optimum storage conditions. Medicated dry foams using nitrofurazone and silver sulfadiazine will be prepared, packaged, and stored at various conditions; in vitro release will be compared before and after storage to determine stability with respect to shelf-life. In addition, the foam will be rendered sterile since it is best to use sterile products on open wounds.
CHAPTER II

EXPERIMENTAL

All chemicals and materials and all equipment used in the experimental procedures are listed in Tables I and II respectively.

Preliminary Studies

At the onset, it was assumed that the original formulation (1) could be satisfactorily packaged in plastic bags. Thus, the following formula was manufactured:

Dextran 16.00 g
Sorbitol 10.40 g
Miranol 0.20 g
Water 76.00 g

The dextran was added to the water and heated on a water bath to 70°. The sorbitol solution was then added and the solution poured into a bowl containing the Miranol. Using an electric kitchen mixer, the solution was whipped for ten minutes producing a light, white foam. The foam was then spread onto four Teflon coated metal sheets, each fitted with two 12'' x 1-1/2'' x 1/16'' plexiglas strips for control of uniform spreading. Prior to spreading the foam, a light coat of silicone was applied to the sheets to aid in removal of the final dry foam. The pans were then placed in a horizontal laminar flow hood with an air velocity of 110 ft/min. and turned every fifteen minutes. After one hour of drying, the dry foam could be easily removed from the pans with a rubber spatula. An oven set at 40-45° would allow faster drying, but because temperatures
<table>
<thead>
<tr>
<th>Name</th>
<th>Lot No. or Other Identifying Marks</th>
<th>Manufacturer</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dextran, M.W. 73,200</td>
<td>34C-1350</td>
<td>Sigma Corporation, St. Louis, Mo.</td>
</tr>
<tr>
<td>Sorbo Solution, 70%</td>
<td>605K6</td>
<td>The Ruger Chemical Co., Irvington, N.J.</td>
</tr>
<tr>
<td>Miranol 2MCA Modified</td>
<td>920SH72ST</td>
<td>The Miranol Co., Irvington, N.J.</td>
</tr>
<tr>
<td>Glycerin</td>
<td>3022002</td>
<td>Colgate-Palmolive Co., New York, N.Y.</td>
</tr>
<tr>
<td>Silicone, D.C. 555</td>
<td>6</td>
<td>The Ruger Chemical Co., Irvington, N.J.</td>
</tr>
<tr>
<td>Nitrofurazone Powder</td>
<td>E9669</td>
<td>Eaton Laboratories, Norwich, N.Y.</td>
</tr>
<tr>
<td>Nitrofurazone Cream, 0.2%</td>
<td>708900</td>
<td>Eaton Laboratories, Norwich, N.Y.</td>
</tr>
<tr>
<td>Silver Sulfadiazine Powder</td>
<td>7115 BS</td>
<td>Marion Laboratories, Kansas City, Mo.</td>
</tr>
<tr>
<td>Silver Sulfadiazine Cream, 1%</td>
<td>E4547</td>
<td>Marion Laboratories, Kansas City, Mo.</td>
</tr>
<tr>
<td>Unibase Cream</td>
<td>PK 310</td>
<td>Parke-Davis Co., Detroit, Mich.</td>
</tr>
<tr>
<td>Calcium Chloride</td>
<td>Drierite</td>
<td>W.A. Hammond Drierite Co., Xenia, Ohio</td>
</tr>
<tr>
<td>Ethylene Oxide, 100%</td>
<td>EPA #7128-1</td>
<td>3M Co., St. Paul, Minn.</td>
</tr>
<tr>
<td>B. subtilis spores</td>
<td>Attest</td>
<td>3M Co., St. Paul, Minn.</td>
</tr>
<tr>
<td>Ethylene Oxide Indicator</td>
<td>Index</td>
<td>3M Co., St. Paul, Minn.</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>A.T.C.C. #25293</td>
<td>American Type Culture Collection, Rockville, Md.</td>
</tr>
<tr>
<td>Antibiotic Medium 2</td>
<td>525075</td>
<td>Difco Laboratories, Detroit, Mich.</td>
</tr>
<tr>
<td>Fluid Thioglycollate Medium</td>
<td>575191</td>
<td>Difco Laboratories, Detroit, Mich.</td>
</tr>
<tr>
<td>Soybean Casein Digest Medium</td>
<td>614363</td>
<td>Difco Laboratories, Detroit, Mich.</td>
</tr>
</tbody>
</table>
TABLE II
Equipment Used in the Experimental Procedures

<table>
<thead>
<tr>
<th>Equipment</th>
<th>Manufacturer</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seal-A-Meal Bags</td>
<td>Dazey Corporation, Kansas City, Mo.</td>
</tr>
<tr>
<td>Heat Sealer</td>
<td>Dazey Corporation, Kansas City, Mo.</td>
</tr>
<tr>
<td>Laminated Aluminum Foil Bags</td>
<td>Eli Lilly Co., Indianapolis, Ind.</td>
</tr>
<tr>
<td>Laminated Aluminum Foil Bags</td>
<td>The Rexham Corporation, Charlotte, N.C.</td>
</tr>
<tr>
<td>Ethylene Oxide Chamber, Model 200</td>
<td>3M Co., St. Paul, Minn.</td>
</tr>
<tr>
<td>Ethylene Oxide Aeration Chamber, Model 33</td>
<td>3M Co., St. Paul, Minn.</td>
</tr>
<tr>
<td>Airguide #605 Relative Humidity Indicator</td>
<td>Airguide Instrument Co., Chicago, Ill.</td>
</tr>
<tr>
<td>Springfield #552 Relative Humidity Indicator</td>
<td>Springfield Instrument Co., Hackensack, N.J.</td>
</tr>
</tbody>
</table>
in an oven are not uniform, uneven drying would occur causing the foam to be too dry in some areas and still be wet in others. Small strips approximately 50 x 30 mm were cut, weighed on an analytical balance, packaged in Seal-A-Meal Bags, and heat sealed. The packaged samples were then stored in a refrigerator (5°; R.H., 55-60%), a freezer (-5°; R.H., 75-90%), and at room conditions (22°; R.H., 40-50%). On the day of manufacture, moisture content was determined by weighing six samples on the analytical balance, drying them to constant weight at ninety volts on the Cenco Moisture Balance, and then calculating moisture content according to the loss in weight. It was assumed that any loss in weight was due only to water loss. Average moisture content on the day of manufacture was found to be 12.7%. Four samples were removed and analyzed for moisture content after one, two, three, four, six, eight, and twelve weeks of storage. Table III summarizes the results of the above procedure. The changes in moisture content indicated that the plastic bags were permeable to moisture and showed that changes in relative humidity could cause the dry foam either to lose or to gain moisture. Maintaining approximately a 10% moisture content is advisable to yield a flexible, nonsticky product (1). That is, a significant loss of moisture causes the foam to lose flexibility while a significant gain causes the foam to become so moist that it adheres to the package or dissolves when touched. The data in Table III indicate the variability of moisture contents of the dry foam packaged in plastic. These changes not only show that plastic alone is not a suitable package, but also confirms the necessity for a moisture impermeable package. Further, the data indicate the need to establish optimum storage conditions with respect to temperature and humidity.
TABLE III

Average Per Cent Moisture Content* of Unmedicated Dry Foam in Plastic Bags

<table>
<thead>
<tr>
<th>Duration of Storage</th>
<th>Room Conditions (22°; R.H., 40-50%)</th>
<th>Refrigerator (+5°; R.H., 55-60%)</th>
<th>Freezer (-5°; R.H., 75-90%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 week</td>
<td>12.6</td>
<td>14.2</td>
<td>16.2</td>
</tr>
<tr>
<td>2 weeks</td>
<td>13.9</td>
<td>14.6</td>
<td>17.6</td>
</tr>
<tr>
<td>3 weeks</td>
<td>14.0</td>
<td>14.5</td>
<td>Deteriorated</td>
</tr>
<tr>
<td>4 weeks</td>
<td>12.6</td>
<td>13.2</td>
<td>Deteriorated</td>
</tr>
<tr>
<td>6 weeks</td>
<td>13.4</td>
<td>12.8</td>
<td>Deteriorated</td>
</tr>
<tr>
<td>8 weeks</td>
<td>11.3</td>
<td>12.2</td>
<td>Deteriorated</td>
</tr>
<tr>
<td>12 weeks</td>
<td>12.7</td>
<td>16.0</td>
<td>Deteriorated</td>
</tr>
<tr>
<td><strong>MEAN</strong></td>
<td><strong>13.0</strong></td>
<td><strong>16.3</strong></td>
<td><strong>16.9</strong></td>
</tr>
</tbody>
</table>

* Average moisture content of 6 samples on day of preparation was 12.7%
Acceptability of a packaging material, based on changes of moisture content in percent, was arbitrarily set as below.

1. Excellent: < 0.5% change; acceptable
2. Good: < 1.0% but > 0.5% change; poss. acceptable
3. Fair: < 2.0% but > 1.0% change; not acceptable
4. Poor: > 2.0% change; not acceptable

Lining the plastic bags with waxed and parchment papers, as well as using double plastic bags, did not improve physical stability of the foam (see Table IV). Rather than search for a moisture impermeable plastic, it was decided to seek alternate packaging materials. Correspondence with a number of packaging manufacturers and users suggested that laminated aluminum foil bags would meet the moisture impermeable requirements most satisfactorily. It was also decided to modify the formula in an attempt to decrease moisture loss by adjusting humectant content.

Laminated aluminum foil bags were secured from Eli Lilly and Co. These bags had a 0.00075 inch thick aluminum foil sandwiched between an outer coat of cellophane and an inner coat of high density polyethylene.

As implied earlier, ideally, the dry foam should be flexible, easy to handle, i.e., does not stick to package or to hands, and be soluble on moist skin. These parameters apply at the day of manufacture as well as after storage at various temperatures and humidities. A lesser but nevertheless significant criterion is the pore size. The dry foam is a very porous formulation and any changes in pore size after storage would indicate deterioration of physical structure.

Nitrofurazone 1% and silver sulfadiazine 1% dry foams were prepared using three different concentrations of glycerin in addition to
<table>
<thead>
<tr>
<th>Duration of Storage</th>
<th>Room Conditions (22°; R.H., 40-50%)</th>
<th>Refrigerator (+5°; R.H., 55-60%)</th>
<th>Freezer (-5°; R.H., 75-90%)</th>
<th>Oven (37°; R.H., 25%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>B</td>
<td>C</td>
<td>A</td>
<td>B</td>
</tr>
<tr>
<td>1 week</td>
<td>6.4% 6.7% 7.1%</td>
<td>8.5% 8.1% 8.9%</td>
<td>9.3% 8.0% 9.4%</td>
<td>4.4% 4.3% 4.7%</td>
</tr>
<tr>
<td>2 weeks</td>
<td>6.3% 6.0% 6.3%</td>
<td>9.0% 8.6% 9.3%</td>
<td>10.6% 8.9% 10.5%</td>
<td>3.1% 3.3% 3.3%</td>
</tr>
<tr>
<td>3 weeks</td>
<td>7.5% 7.3% 7.3%</td>
<td>9.2% 9.1% 9.2%</td>
<td>12.6% 10.5% 11.6%</td>
<td>5.2% 5.2% 5.3%</td>
</tr>
<tr>
<td>4 weeks</td>
<td>6.6% 6.4% 6.7%</td>
<td>9.6% 9.2% 9.4%</td>
<td>15.3% 12.8% 13.8%</td>
<td>3.9% 3.9% 3.9%</td>
</tr>
<tr>
<td>MEAN</td>
<td>6.7% 6.7% 6.9%</td>
<td>9.1% 8.8% 9.2%</td>
<td>12.0% 10.1% 11.3%</td>
<td>4.2% 4.2% 4.3%</td>
</tr>
</tbody>
</table>

A = bags lined with waxed paper; average moisture content of 12 samples on day of preparation = 9.3%
B = bags lined with parchment paper; ave. moisture content of 12 samples on day of preparation = 9.0%
C = double bagged; average moisture content of 12 samples on day of preparation = 10.1%
sorbitol. Also, the original formula without glycerin was manufactured for comparison. Moisture content and subjective evaluation of gross physical properties, i.e., flexibility, stickiness, dissolution time, porosity, were determined on the day of manufacture. Samples of each dry foam were then weighed, packaged in the foil bags, and heat sealed. Three samples were stored for one week at the following conditions:

1. Freezer (-5°; R.H., 75-90%)
2. Dessicator (22°; R.H., 5-6%)

The various formulas for nitrofurazone dry foams are listed in Table V and for silver sulfadiazine, Table VI. Table VII lists the moisture content and subjective evaluation of the nitrofurazone formulas on the day of manufacture and after storage. Table VIII summarizes these data for silver sulfadiazine foam.

Because these formulas (see Tables V and VI) were too sticky, three additional formulas with decreased concentrations of glycerin were prepared and compared to the original. Additional storage conditions of room temperature, refrigerator, and a laboratory oven were included in the stability studies. Because laminated aluminum foil bags maintained satisfactory moisture content (see Table VII and VIII), moisture content determination was not done in this part of the study. The additional formulas are listed in Table IX for nitrofurazone and Table X for silver sulfadiazine. The subjective evaluations are summarized in Tables XI and XII respectively.

These preliminary tests indicated that modification of the original formula with glycerin in concentrations greater than 0.19% wet weight, i.e., formulas II-VI of nitrofurazone and silver sulfadiazine,
TABLE V
Composition of Original (I) and Modified Formulas (II-IV) of 1% Nitrofurazone Dry Foam

<table>
<thead>
<tr>
<th>Formula Number</th>
<th>Water</th>
<th>Dextran</th>
<th>Sorbitol</th>
<th>Glycerin</th>
<th>Miranol</th>
<th>Nitrofurazone</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>28.5*</td>
<td>6.0</td>
<td>3.9</td>
<td></td>
<td>0.075</td>
<td>0.10</td>
</tr>
<tr>
<td>II</td>
<td>27.6</td>
<td>6.0</td>
<td>3.9</td>
<td>0.94</td>
<td>0.075</td>
<td>0.11</td>
</tr>
<tr>
<td>III</td>
<td>27.8</td>
<td>6.0</td>
<td>3.9</td>
<td>0.75</td>
<td>0.075</td>
<td>0.11</td>
</tr>
<tr>
<td>IV</td>
<td>28.1</td>
<td>6.0</td>
<td>3.9</td>
<td>0.38</td>
<td>0.075</td>
<td>0.11</td>
</tr>
</tbody>
</table>

* Quantities are listed in grams.

TABLE VI
Composition of Original (I) and Modified Formulas (II-IV) of 1% Silver Sulfadiazine Dry Foam

<table>
<thead>
<tr>
<th>Formula Number</th>
<th>Water</th>
<th>Dextran</th>
<th>Sorbitol</th>
<th>Glycerin</th>
<th>Miranol</th>
<th>Ag Sulfadiazine</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>28.5*</td>
<td>6.0</td>
<td>3.9</td>
<td></td>
<td>0.075</td>
<td>0.10</td>
</tr>
<tr>
<td>II</td>
<td>27.6</td>
<td>6.0</td>
<td>3.9</td>
<td>0.94</td>
<td>0.075</td>
<td>0.11</td>
</tr>
<tr>
<td>III</td>
<td>27.8</td>
<td>6.0</td>
<td>3.9</td>
<td>0.75</td>
<td>0.075</td>
<td>0.11</td>
</tr>
<tr>
<td>IV</td>
<td>28.1</td>
<td>6.0</td>
<td>3.9</td>
<td>0.38</td>
<td>0.075</td>
<td>0.11</td>
</tr>
</tbody>
</table>

* Quantities are listed in grams,
TABLE VII
Average Per Cent Moisture Content and Gross Physical Appearance (GPA) of Nitrofurazone Formulas in Foil Bags* Before and After Storage

<table>
<thead>
<tr>
<th>Formula No.</th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>GPA on Day of Preparation</th>
<th>GPA in Freezer ((-5^\circ; \text{R.H. 75-90%}))</th>
<th>GPA in Dessicator ((22^\circ; \text{R.H. 5-6%}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>10.0%</td>
<td>10.8%</td>
<td>10.2%</td>
<td>Flexible (\text{Dissolved on moist skin in 15 sec.}) Not sticky Even porosity</td>
<td>Unchanged</td>
<td>Stuck to bags Slight loss of flexibility Other properties unchanged</td>
</tr>
<tr>
<td>II</td>
<td>9.8%</td>
<td>9.8%</td>
<td>10.1%</td>
<td>Very flexible (\text{Dissolved on moist skin in 15 sec.}) Sticky Even porosity</td>
<td>Increased porosity Other properties unchanged</td>
<td>Great increase in porosity Stuck to bags Other properties unchanged</td>
</tr>
<tr>
<td>III</td>
<td>9.1%</td>
<td>8.9%</td>
<td>9.3%</td>
<td>Same as II</td>
<td>Same as II</td>
<td>Same as II</td>
</tr>
<tr>
<td>IV</td>
<td>9.3%</td>
<td>9.0%</td>
<td>9.4%</td>
<td>Less sticky than II and III Other properties same as II &amp; III</td>
<td>Unchanged</td>
<td>Slightly stuck to bags Slight loss of flexibility Other properties unchanged</td>
</tr>
</tbody>
</table>

* Eli Lilly and Co., Indianapolis, Indiana
TABLE VIII

Average Per Cent Moisture Content and Gross Physical Appearance (GPA) of Silver Sulfadiazine Formulas in Foil Bags*·Before-and·After Storage

<table>
<thead>
<tr>
<th>Formula No.</th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>GPA on Day of Preparation</th>
<th>GPA in Freezer  (-5^\circ;) R.H. 75-90%</th>
<th>GPA in Dessicator (22^\circ;) R.H. 5-6%</th>
</tr>
</thead>
</table>
| I          | 9.8% | 10.9%| 9.4% | Flexible  
Dissolved on moist skin in 15 sec.  
Not sticky  
Even porosity | Unchanged | Slightly stuck to bags  
Increased porosity  
Slight loss of flexibility  
Other properties unchanged |
| II         | 10.5%| 10.4%| 9.7% | Very flexible  
Dissolved on moist skin in 15 sec.  
Sticky  
Even porosity | Increased porosity  
Increased stickiness  
Other properties unchanged | Great increase in porosity  
Stuck to bags  
Loss of flexibility  
Other properties unchanged |
| III        | 9.3% | 9.0% | 9.1% | Same as II  
Even porosity  
Other properties unchanged | Same as II | Same as II |
| IV         | 8.8% | 8.8% | 8.7% | Less sticky than II and III  
Other properties same as II & III | Unchanged | Same as I in dess. except still sticky |

* Eli Lilly and Co., Indianapolis, Indiana
### TABLE IX

Composition of Original (I) and Additional Formulas (V-VII) of 1% Nitrofurazone Dry Foam

<table>
<thead>
<tr>
<th>Formula Number</th>
<th>Water</th>
<th>Dextran</th>
<th>Sorbitol</th>
<th>Glycerin</th>
<th>Miranol</th>
<th>Nitrofurazone</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>28.5*</td>
<td>6.0</td>
<td>3.9</td>
<td></td>
<td>0.075</td>
<td>0.10</td>
</tr>
<tr>
<td>V</td>
<td>28.2</td>
<td>6.0</td>
<td>3.9</td>
<td>0.30</td>
<td>0.075</td>
<td>0.10</td>
</tr>
<tr>
<td>VI</td>
<td>28.4</td>
<td>6.0</td>
<td>3.9</td>
<td>0.15</td>
<td>0.075</td>
<td>0.10</td>
</tr>
<tr>
<td>VII</td>
<td>28.4</td>
<td>6.0</td>
<td>3.9</td>
<td>0.075</td>
<td>0.075</td>
<td>0.10</td>
</tr>
</tbody>
</table>

* Quantities are listed in grams

### TABLE X

Composition of Original (I) and Additional Formulas (V-VII) of 1% Silver Sulfadiazine Dry Foam

<table>
<thead>
<tr>
<th>Formula Number</th>
<th>Water</th>
<th>Dextran</th>
<th>Sorbitol</th>
<th>Glycerin</th>
<th>Miranol</th>
<th>Ag Sulfadiazine</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>28.5*</td>
<td>6.0</td>
<td>3.9</td>
<td></td>
<td>0.075</td>
<td>0.10</td>
</tr>
<tr>
<td>V</td>
<td>28.2</td>
<td>6.0</td>
<td>3.9</td>
<td>0.30</td>
<td>0.075</td>
<td>0.10</td>
</tr>
<tr>
<td>VI</td>
<td>28.4</td>
<td>6.0</td>
<td>3.9</td>
<td>0.15</td>
<td>0.075</td>
<td>0.10</td>
</tr>
<tr>
<td>VII</td>
<td>28.4</td>
<td>6.0</td>
<td>3.9</td>
<td>0.075</td>
<td>0.075</td>
<td>0.10</td>
</tr>
</tbody>
</table>

* Quantities are listed in grams
<table>
<thead>
<tr>
<th>Formula No.</th>
<th>GPA on Day of Preparation</th>
<th>Gross Physical Appearance After Storage in</th>
<th>Freezer (-5°; R.H. 75-90%)</th>
<th>Refrigerator (+5°; R.H. 55-60%)</th>
<th>Dessicator (22°; R.H. 5-6%)</th>
<th>Oven (37°; R.H. 25%)</th>
<th>Room (22°; R.H. 45%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Flexible Dissolved on moist skin in 15 sec. Not sticky Even porosity</td>
<td></td>
<td>Increased porosity Other properties unchanged</td>
<td>Unchanged</td>
<td>Loss of flexibility Other properties unchanged</td>
<td>Totally deteriorated</td>
<td>Same as I in dessicator</td>
</tr>
<tr>
<td>V</td>
<td>Flexible Dissolved on moist skin in 15 sec. Less sticky than II-IV Even porosity</td>
<td></td>
<td>Increased stickiness Slight increase in porosity Other properties unchanged</td>
<td>Same as V in freezer</td>
<td>Loss of flexibility Great increase in porosity Stuck to bags Other properties unchanged</td>
<td>Totally deteriorated</td>
<td>Same as V in dessicator</td>
</tr>
<tr>
<td>VI</td>
<td>Very flexible Dissolved on moist skin in 15 sec. Less sticky than II-V Even porosity</td>
<td></td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Same as V in dessicator</td>
<td>Totally deteriorated</td>
<td>Same as V at room</td>
</tr>
<tr>
<td>VII</td>
<td>Very flexible Dissolved on moist skin in 15 sec. Not sticky Even porosity</td>
<td></td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Very slight loss of flexibility Other properties unchanged</td>
<td>Totally deteriorated</td>
<td>Same as VII in dessicator</td>
</tr>
</tbody>
</table>
### TABLE XII

Gross Physical Appearance (GPA) of Additional Silver SulfaDiazine Formulas Before and After Storage

<table>
<thead>
<tr>
<th>Formula No.</th>
<th>GPA on Day of Preparation</th>
<th>Gross Physical Appearance After Storage in</th>
<th>Freezer (-5°C; R.H. 75-90%)</th>
<th>Refrigerator (+5°C; R.H. 55-60%)</th>
<th>Dessicator (220°C; R.H. 5-6%)</th>
<th>Oven (37°C; R.H. 25%)</th>
<th>Room (220°C; R.H. 45%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Flexible</td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Slight loss of flexibility Other properties unchanged</td>
<td>Totally deteriorated</td>
<td>Unchanged</td>
</tr>
<tr>
<td></td>
<td>Dissolved on moist skin in 15 sec. Not sticky Even porosity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>V</td>
<td>Very flexible</td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Loss of flexibility Great increase in porosity Other properties unchanged</td>
<td>Totally deteriorated</td>
<td>Unchanged</td>
</tr>
<tr>
<td></td>
<td>Dissolved on moist skin in 15 sec. Less sticky than II-IV Even porosity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VI</td>
<td>Very flexible</td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Same as V in dessicator</td>
<td>Totally deteriorated</td>
<td>Same as V in dessicator</td>
</tr>
<tr>
<td></td>
<td>Dissolved on moist skin in 15 sec. Less sticky than II-V Even porosity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VII</td>
<td>Very flexible</td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Unchanged</td>
<td>Same as I in dessicator</td>
<td>Totally deteriorated</td>
<td>Unchanged</td>
</tr>
<tr>
<td></td>
<td>Dissolved on moist skin in 15 sec. Not sticky Even porosity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
caused the dry foam to be too sticky such that it could not be easily handled. In a concentration of 0.19%, i.e., formula VII, the dry foam was very similar to the original, but because it was felt that glycerin would decrease the tendency to lose moisture and thus, increase the chances of maintaining flexibility, formula VII was chosen for further evaluation.

From these preliminary tests, a three month storage study was conducted using formula VII for nitrofurazone and silver sulfadiazine dry foams packaged in foil laminates and stored in a freezer, refrigerator, dessicator, and at room temperature. Storage at elevated temperatures much greater than room temperature were not satisfactory, regardless of the package or the formula. Also, the preliminary studies indicated that laminated aluminum foil bags fulfilled the moisture impermeable requirements (see Tables XI and XII). However, because of the unavailability of the original foil bags, another type had to be obtained. This change was not considered significant since all laminated aluminum foils are considered impermeable to moisture. Thus, different laminated aluminum foil bags were secured from the Rexham Corporation; these new bags, starting from the inner side, consisted of a paper/primer/polyethylene/.001" aluminum foil/vinyl wash/resin coating.

Procedure for Nitrofurazone 1% Dry Foam

The following formula was prepared:

- Dextran 16.00 g
- Sorbitol 10.40 g
- Glycerin 0.20 g
- Miranol 0.20 g
Nitrofurazone 0.27 g
Water 75.80 g

The identical procedures previously described for manufacturing, moisture content determination, and packaging were followed. Calculations for determining the amount of active ingredient needed were done on a dry weight basis using 10% as the average moisture content. The average moisture content of ten samples on the day of manufacture was found to be 9.7%.

A drug product must be both physically and therapeutically stable. Moisture content mainly affects the physical properties of the dry foam; to determine if storage conditions, packaging material, and/or time affected the therapeutic efficacy, in vitro release using a modified agar plate method was done. Antibiotic Medium 2 was made according to the manufacturer's directions, autoclaved at 15 psi and 121° for 30 minutes. Twenty-five ml were then poured into 15 x 100 mm disposable petri dishes. The agar was allowed to gel and ten plates were seeded with 0.1 ml of an overnight inoculum of Staphylococcus aureus A.T.C.C. #25293. Two sterile penicylinders were placed on the gelled agar plates; one was filled with approximately 0.3 g of Furacin Cream 0.2% and the other, with Unibase to serve as a control. One disc of the freshly made dry nitrofurazone foam was placed on each of the ten plates. The plates were incubated at 37° for 24 hours following which the zones of inhibition were read, using the Fisher-Lilly Zone Reader.

At monthly intervals, for a period of three months, four samples of the dry foam were removed from each storage condition and moisture content determined. Also, one sample from each condition was removed and in vitro release was determined using the above procedure.
Preliminary work with nitrofurazone indicated that 0.2% nitrofurazone dry foam did not give equal or greater zones of inhibition than the commercial cream. Probably, this was not because the foam did not release the drug, but simply because there was much less nitrofurazone, i.e., subtherapeutic concentrations, per disc of foam than in 0.3 g of Furacin Cream. Therefore, the concentration of nitrofurazone in the foam had to be increased to provide therapeutic concentrations. Table XIII summarizes the average zones of inhibition from ten readings obtained with varying concentrations of nitrofurazone. One percent was selected as therapeutically equivalent to the commercial cream.

Results for nitrofurazone dry foam for moisture content and in vitro release on the day of manufacture and at one month intervals of storage are listed in Tables XIV and XV. Each sample shown in the tables was taken from its own foil bag.

Procedure for Silver Sulfadiazine 1% Dry Foam

The following formula was manufactured:

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dextran</td>
<td>16.00 g</td>
</tr>
<tr>
<td>Sorbitol</td>
<td>10.40 g</td>
</tr>
<tr>
<td>Glycerin</td>
<td>0.20 g</td>
</tr>
<tr>
<td>Miranol</td>
<td>0.20 g</td>
</tr>
<tr>
<td>Silver sulfadiazine</td>
<td>0.27 g</td>
</tr>
<tr>
<td>Water</td>
<td>75.80 g</td>
</tr>
</tbody>
</table>

The identical procedure described for nitrofurazone dry foam was employed for silver sulfadiazine with respect to preparation, moisture content calculations, and determination of a suitable package and of proper storage conditions. Average moisture content of ten samples of silver
TABLE XIII
Zones of Inhibition Obtained with Various Concentrations of Nitrofurazone

<table>
<thead>
<tr>
<th>Drug</th>
<th>Mean Diameter (mm) (average of 10 readings)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Furacin Cream 0.2%</td>
<td>12.1</td>
</tr>
<tr>
<td>Nitrofurazone Foam 0.2%</td>
<td>not distinguishable</td>
</tr>
<tr>
<td>Nitrofurazone Foam 0.5%</td>
<td>6.4</td>
</tr>
<tr>
<td>Nitrofurazone Foam 0.75%</td>
<td>10.9</td>
</tr>
<tr>
<td>Nitrofurazone Foam 1.0%</td>
<td>12.9</td>
</tr>
<tr>
<td>Nitrofurazone Foam 2.0%</td>
<td>14.3</td>
</tr>
<tr>
<td>Nitrofurazone Foam 3.0%</td>
<td>19.7</td>
</tr>
<tr>
<td>Control*</td>
<td>0.0</td>
</tr>
</tbody>
</table>

* Unibase, Parke-Davis Co., Detroit, Mich.
## TABLE XIV

Moisture Content (%) of 1% Nitrofurazone Dry Foam

on the Day of Preparation and After Storage

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Day of Preparation</th>
<th>One Month's Storage</th>
<th>Two Months' Storage</th>
<th>Three Months' Storage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>B</td>
<td>C</td>
</tr>
<tr>
<td>1</td>
<td>9.8</td>
<td>8.8</td>
<td>9.2</td>
<td>9.6</td>
</tr>
<tr>
<td>2</td>
<td>10.2</td>
<td>10.8</td>
<td>9.7</td>
<td>9.3</td>
</tr>
<tr>
<td>3</td>
<td>9.6</td>
<td>8.6</td>
<td>10.1</td>
<td>9.1</td>
</tr>
<tr>
<td>4</td>
<td>10.8</td>
<td>9.6</td>
<td>9.4</td>
<td>9.7</td>
</tr>
<tr>
<td>5</td>
<td>10.4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>9.3</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>8.8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>9.4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>8.8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>9.8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MEAN</td>
<td>9.7</td>
<td>9.5</td>
<td>9.4</td>
<td>9.7</td>
</tr>
<tr>
<td>p&lt;sup&gt;2&lt;/sup&gt;</td>
<td>&lt;.7</td>
<td>&lt;.9</td>
<td>&lt;.5</td>
<td>&lt;.001</td>
</tr>
</tbody>
</table>

A = 22°; R.H., 47-51%
B = +5°; R.H., 55-60%
C = -5°; R.H., 75-90%
D = 22°; R.H., 5-6%

p<sup>2</sup> Student t calculations comparing moisture content on day of preparation with moisture content after storage
### TABLE XV

Diameters (mm) of Zones of Inhibition of 0.2% Nitrofurazone Cream and 1% Nitrofurazone Dry Foam on the Day of Preparation and After Storage

<table>
<thead>
<tr>
<th>Plate No.</th>
<th>Day of Preparation</th>
<th>One Month</th>
<th>Two Months</th>
<th>Three Months</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cr Foam</td>
<td>Cr A B C D</td>
<td>Cr A B C D</td>
<td>Cr A B C D</td>
</tr>
<tr>
<td>1</td>
<td>11.9</td>
<td>9.4 11.2 10.7 10.0 10.1</td>
<td>9.3 11.7 10.7 12.6 9.6</td>
<td>9.8 13.1 11.4 12.8 12.0</td>
</tr>
<tr>
<td>2</td>
<td>9.0 9.5 8.6 9.6 11.3 12.9 9.7</td>
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A = 22°; R.H., 47-51%
B = +5°; R.H., 55-60%
C = -5°; R.H., 75-90%
D = 22°; R.H., 5-6%

- P<sub>1</sub> - Student t; cream vs. foam on day of preparation
- P<sub>2</sub> - Student t; cream vs. cream after storage
- P<sub>3</sub> - Student t; foam vs. foam after storage
sulfadiazine dry foam on the day of manufacture was found to be 9.8%. In vitro release was compared to the commercially available silver sulfadiazine 1% cream.

Results for silver sulfadiazine dry foam for moisture content and in vitro release on the day of manufacture and at monthly intervals of storage are listed in Tables XVI and XVII. As with nitrofurazone, each sample shown in the tables for silver sulfadiazine was taken from its own foil bag.

Sterilization Procedures

**Method I:**

Unmedicated casting solution, i.e., dextran, sorbitol, glycerin, Miranol, and water, was inoculated with 0.2 ml of an overnight inoculum of *S. aureus* and then autoclaved at 15 psi and 121° for 20 minutes. The resultant solution was placed in a sterile bowl in a laminar flow hood and whipped with sterile beaters. The whipped foam was aseptically spread onto sterilized pans and allowed to dry in the hood.

**Method II:**

Unmedicated casting solution inoculated with 0.2 ml of an overnight inoculum of *S. aureus* was filtered through a 0.22 micron Millipore filter into a sterile bowl. This was done in a laminar flow hood using a 50 ml syringe. The solution was whipped with sterile beaters and aseptically spread onto sterile pans and allowed to dry in the hood.

**Method III:**

Unmedicated casting solution was inoculated with 0.2 ml of an
**TABLE XVI**

Moisture Content (%) of 1% Silver Sulfadiazine Dry Foam on the Day of Preparation and After Storage

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Day of Preparation</th>
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<th>Two Months' Storage</th>
<th>Three Months' Storage</th>
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A = 22°; R.H., 47-51%<br>
B = +5°; R.H., 55-60%<br>
C = -5°; R.H., 75-90%<br>
D = 22°; R.H., 5-6%<br>

*p* Student t calculations comparing moisture content on day of preparation with moisture content after storage.
TABLE XVII

Diameters (mm) of Zones of Inhibition of 1% Silver Sulfadiazine Cream and
1% Silver Sulfadiazine Dry Foam on the Day of Preparation and After Storage

<table>
<thead>
<tr>
<th>Plate No.</th>
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A = 22°; R.H., 47-51%
B = +5°; R.H., 55-60%
C = -5°; R.H., 75-90%
D = 22°; R.H., 5-5%

Student t; foam vs. foam on day of preparation

Student t; foam vs. foam after storage

Student t; cream vs. foam after storage
overnight inoculum of \textit{S. aureus} and whipped in a bowl. The whipped foam was spread onto a pan and allowed to dry in a laminar flow hood. The dry foam was removed and placed in ethylene oxide permeable plastic bags obtained from Tower Products, Inc. The packaged foam was then placed in a cold cycle (29°) ethylene oxide chamber and the cycle allowed to run for three hours using 100% ethylene oxide. Spores of \textit{B. subtilis} were used as the biological indicator while Indox Ethylene Oxide Indicator served as the chemical indicator. Upon completion of the cycle, both indicators showed that ethylene oxide sterilization requirements were met. The packaged foam was then aerated for eight hours at 63° in an aeration chamber to allow dissipation of ethylene oxide residues.

Method IV:

Unmedicated casting solution was inoculated with 0.2 ml of an overnight inoculum of \textit{S. aureus}. The solution was whipped, spread onto pans, and allowed to dry as above. The contaminated dry foam was packaged in aluminum foil laminates provided by the Rexham Corporation and mailed to International Nutronics, Inc., a commercial radiation sterilizing company. Cobalt-60 was the source of the gamma radiation using a total dose of 2.5 Mrads. After sterilization, the packages were returned unopened to the laboratory for testing.

Sterility Testing Procedures:

Samples of dry foams sterilized by the four methods above were tested for sterility. The USP XVIII method using Fluid Thioglycollate
Medium (FTM) and Soybean Casein Digest Medium (SCD) was employed. To two test tubes each of FTM and SCD were added samples from each method. The test tubes were allowed to incubate in an incubating oven for 14 days at 37°. Also, two positive controls, i.e., tubes of media containing 0.1 ml of an overnight inoculum of *S. aureus*, and two negative controls were used for comparison. Samples of the contaminated dry foams prior to sterilization by ethylene oxide and radiation were added to additional test tubes of FTM and SCD and incubated with the other test media.

Samples of the two aseptic preparation methods (Methods I and II) were packaged in laminated foil bags and stored at room conditions for one month. The samples were then aseptically removed and tested for sterility.

**Miscellaneous Procedures:**

The most common defect with laminated aluminum bags is an ineffective seal. Thus, fifty bags were sealed with a heat sealer and totally immersed in water for twenty-four hours. Upon removal and opening of the bags, none contained moisture indicating that the sealing procedure was effective.

To determine if the dry foam could withstand normal shipping and mailing conditions, four samples of unmedicated, uncontaminated dry foam were packaged in laminated foil and shipped by mail to the East coast and returned unopened to the laboratory. The same packages were sent immediately to a second West coast city and again returned unopened. These shipped samples were then compared to control samples from the same batch that were packaged and kept in the laboratory.
CHAPTER III

RESULTS

Nitrofurazone Stability

Table XIV summarizes the average moisture content of packaged nitrofurazone dry foam on the day of manufacture and after one, two, and three months' storage at four different storage conditions. As shown, the average moisture content of ten samples on the day of manufacture was 9.7%.

After one month's storage in a refrigerator (+5°; R.H., 55-60%), freezer (-5°; R.H., 75-90%), and at room conditions (22°; R.H., 50%) the changes in moisture content were statistically insignificant with the largest change, i.e., 9.7% to 9.4%, occurring in the freezer. The insignificance of these small changes in moisture is not only shown statistically, but also reflected by the fact that, in all cases, the nitrofurazone dry foam retained its flexibility, porosity, and nonstickiness. However, when stored in a dessicator (22°; R.H., 5-6%) for one month, the moisture content dropped from 9.7% to 7.1% (P < .001) and the foam became very friable.

After two months' storage in a refrigerator and at room conditions with an average relative humidity of 47%, no significant changes in moisture content and gross physical properties were noted. A drop to 9.5% moisture content occurred in the freezer, but again, this was negligible as the foam still possessed the ideal characteristics. In the dessicator, the moisture content dropped from 9.7% on the day of
manufacture to 8.4% ($P > .02 < .05$) causing the foam to lose its flexibility.

No changes in physical properties occurred after three months' storage in the refrigerator, freezer, and at room conditions (R.H., 48%) despite the fact that the moisture content of the foam in the freezer and at room temperature increased to 9.9% while it decreased to 9.2% in the refrigerator. These changes are statistically insignificant. Again, the moisture content of the foam dropped to 7.7% ($P > .01 < .02$) in the dessicator with subsequent loss of flexibility.

Table XV lists the zones of inhibition obtained with nitrofurazone dry foam on the day of manufacture and after one, two, and three months' storage at each of the four storage conditions. Also included in the table are the zones of inhibition obtained with the commercially available nitrofurazone cream. Storage of the nitrofurazone dry foam at room conditions, a freezer, and a dessicator for one and two months did not produce statistically different zones of inhibition from the zones produced on the day of manufacture. Samples stored in a refrigerator for one and two months did, however, produce statistically different zones as did storage for three months at all four conditions. These statistically significant diameters of zones after storage were all greater than those on the day of manufacture.

Silver Sulfadiazine Stability

Table XVI summarizes the average moisture content of packaged silver sulfadiazine dry foam on the day of manufacture and after one, two, and three months' storage at the four different storage conditions. The average moisture content from ten readings on the day of manufacture
A drop in moisture content from 9.8% to 9.0% was noted in the samples stored at room conditions for one month; a slight increase in porosity occurred, but flexibility was still maintained. In the refrigerator and freezer after one month, the silver sulfadiazine samples all retained their original physical properties despite slight changes in moisture content. The samples stored in the dessicator for one month demonstrated a loss of moisture content from 9.8% to 9.1%; a slight increase in porosity was noted, but flexibility was still maintained. Statistical evaluation showed that these changes were all insignificant.

After two months' storage, all the samples under all the storage conditions retained their original gross physical properties. The samples at room conditions had a drop in moisture content from 9.8% to 9.3% while the samples in the refrigerator, freezer, and dessicator all gained moisture from 9.8% to 10.2%, 10.0%, and 10.6% respectively. Again, these changes were statistically insignificant.

Samples stored for three months at room conditions and in the freezer lost slight amounts of moisture but demonstrated no changes in physical properties. After three months in the refrigerator, again the samples retained their physical qualities despite a slight increase in moisture content to 9.9%. Samples in the dessicator lost slightly more than 1% moisture but still possessed adequate flexibility although they exhibited a relatively large increase in porosity. These moisture content changes were all statistically insignificant.

Table XVII gives the zones of inhibition obtained with silver sulfadiazine dry foam on the day of manufacture and after storage under conditions identical to those for nitrofurazone preparations. The zones
of inhibition given by the commercially available silver sulfadiazine 1% cream are also included in Table XVII. No statistically significant differences were noted between the silver sulfadiazine dry foam zones obtained on the day of manufacture and after one, two, and three months' storage regardless of the storage condition with one exception. After three months at room conditions, a statistically smaller mean zone of inhibition was produced ($P < .001$).

Results of Sterilization of Dry Foam

Method I:

Sterilization of the inoculated casting solution by autoclaving did not produce any changes in its consistency and spreadability during the subsequent aseptic preparation of the dry foam. The final product also did not show any physical changes. Samples of the dry foam so prepared did not show any growth on incubation in the two media used to test their sterility. The positive controls showed growth while the negative controls showed no growth.

Method II:

Filtration of the inoculated casting solution also did not alter the consistency and spreadability of the whipped foam and/or the physical properties of the final product. Aseptic preparation of the dry foam produced a sterile product as no growth occurred after fourteen days' incubation. Both negative controls of each medium also exhibited no growth while the positive controls did.

Samples of dry foam prepared by Methods I and II above and aseptically packaged in laminated aluminum foil were tested for
sterility after storage for one month at room conditions. These samples also showed sterility despite the fact that the insides of the foil packages were only cleaned by wiping with 70% isopropanol (rubbing alcohol).

Method III:

Samples of inoculated dry foam prior to ethylene oxide sterilization showed growth in the media used. Separate samples of foam after ethylene oxide sterilization showed sterility. However, after the sterilization-aeration cycles were completed, the foam showed physical deterioration such as loss of flexibility and increase in porosity.

Method IV:

The irradiation sterilized samples of dry foam also met the test for sterility. Freshly inoculated samples prior to sterilization, as expected, showed growth. Some loss of flexibility of the foam after irradiation sterilization was observed. However, this loss was not significant enough to affect the desirable features of the product.

Results of Shipping

Comparison of the shipped samples and controls kept in the laboratory showed that there was no loss of desirable features of flexibility and porosity on shipping.
CHAPTER IV

DISCUSSION

Conventional topical dosage forms consist of solutions, lotions, creams, ointments, and aerosols. While these types of products are widely used, they are not without some inherent disadvantages such as necessity for removal, waste especially from aerosols, and possible aggravation of open lesions via inunction. Because of these disadvantages, a novel topical dressing which would dissolve on moist skin, release the active ingredient without inunction, and not necessitate removal was developed (1-3). However, this dressing had its own disadvantages; the most significant being loss of moisture and flexibility prior to and during storage. Therefore, the use of a moisture impermeable package and determination of suitable storage conditions were necessary.

Because of the inexpensiveness and great versatility of plastics, a twelve week storage study was designed using what was thought to be a moisture proof plastic bag (see Table III). After twelve weeks of storage in a refrigerator with periodic evaluation of moisture content, it was found that the dry foam gained moisture and became very sticky and difficult to handle. In a freezer, the foam liquified completely in just four weeks. Storage at room conditions produced very inconsistent data. An increase in moisture content occurred after two and three weeks' storage while a decrease occurred after four weeks. Similarly, six weeks' storage showed increased moisture content while eight and twelve weeks' caused decreased moisture content. These room condition
samples maintained good flexibility without acquiring a tacky feeling. The results of this study confirmed that the dry foam was sensitive to moisture and indicated that the plastic bags were permeable to moisture.

Consequently, a four week storage study employing plastic bags lined with waxed-paper and parchment paper as well as double-bagging was initiated to determine if these liners or double thickness bags would minimize moisture permeation (see Table IV). With waxed-paper or double bagging, moisture content dropped sufficiently to cause loss of flexibility in samples stored at room conditions; samples packaged with parchment liner also lost moisture, but flexibility was maintained after one, two, three, and four weeks. All samples stored at 37° lost moisture and flexibility regardless of the type of liner or double bagging, even after only one week of storage. Freezer samples, again regardless of the type of package, gained sufficient moisture after three and four weeks to cause tackiness such that the foam could not be easily handled; samples stored in a refrigerator all maintained adequate flexibility without tackiness after four weeks.

One would expect that if the foam gains or loses moisture at a particular storage condition, such a change would be steady and consistent. That is, if a gain were noted after one week storage, then a drop in moisture after two or three weeks should not occur. However, this was not the case, especially at room conditions. These inconsistent data (see Tables III and IV) indicated that some other factor besides temperature and type of package was also playing an important role in the physical stability of the foam. This factor was finally determined to be relative humidity which, of course, changes from time to time, not only at room conditions, but also in refrigerators and freezers. Although
Tables III and IV indicate relative humidity at the various storage conditions; this was not monitored during these two preliminary studies because it wasn't anticipated that relative humidity would be a significant factor.

These results showed that 1) the plastic bags were permeable, 2) both temperature and relative humidity during storage must be controlled, and 3) the original formula might be improved to produce a more stable dry foam. As stated earlier, a significant drop in moisture content would cause a loss of flexibility while an increase may cause the foam to be too tacky. Of these two possibilities, loss of flexibility is the more important because an inflexible dressing cannot be applied to anatomical areas that have nonplanar geometry. Although a tacky product could be applied to body areas, its handling may require forceps or it may adhere to the package.

The loss of flexibility may be attributed to crystallization of sorbitol occurring on loss of water. Glycerin in varying concentrations was used as a second humectant and an adjunct to maintain sorbitol in solution, resulting in a more pliable foam. Packaging material other than plastic bags would also be required to maintain moisture content of the foam. Laminated aluminum foil was selected since it would be the most satisfactory moisture proof package. Consequently, foil laminates were used during the remainder of the study.

Three different dry foam formulas containing nitrofurazone and silver sulfadiazine were manufactured, packaged in the foil bags, and stored for one week at the two extremes of relative humidity, freezer (R.H., 75-90%) and dessicator (R.H., 5-6%). These modified formulas are listed in Tables V and VI. Moisture content on the day of manufacture
and after storage as well as subjective comments regarding physical properties are summarized in Tables VII and VIII. Comparison of the three modified formulas with the original, showed that formulas II and III became very tacky after one week in the freezer, and exhibited a slight loss in flexibility and an increase in porosity in the dessicator. Formula IV, while less tacky than formulas II and III, was more sticky than the original. After storage in the freezer, the tackiness of formula IV was relatively satisfactory, but not ideal. Both the original formula and formula IV showed slight losses of flexibility and increases in porosity after storage in the dessicator. Changes in physical properties would appear to be directly related to changes in moisture content. In fact, this was the major assumption and reason for finding a moisture impermeable package and for defining exact storage conditions with respect to relative humidity and temperature. However, study of formulas II through IV showed that this was not the entire picture, but that factors other than moisture content also cause physical changes on storage. Formulas II-IV as well as the original formula did not gain or lose significant amounts of moisture when packaged in foil and yet, they exhibited unsatisfactory changes in physical properties. To explain these changes is difficult, but they apparently are inherent in the formula.

Because formulas II-IV exhibited unsatisfactory physical changes, additional formulas, V-VII, with decreasing concentrations of glycerin were prepared. These additional formulas are listed in Tables IX and X. They were evaluated for physical properties on the day of manufacture and then packaged in foil and stored for one week at room conditions, in a freezer, a refrigerator, a dessicator, and an oven set at 37°. Because
the previous study showed that the foil bags were very satisfactory in preventing moisture content changes, the purpose of this work was only to find a more suitable formula that did not possess the unknown, inherent factors alluded to earlier. A more suitable formula would be defined as one yielding a nonsticky, flexible product just as the original but which also contained some glycerin to help maintain flexibility. The tackiness of formulas II-IV was caused by excess glycerin. A formula with the correct concentration of glycerin may not possess this undesirable tackiness while it might retain greater flexibility than the original, nonglycerin formula. Tables XI and XII summarize the physical properties of the original formula and the additional modified formulas V-VII. Formulas V and VI showed similar results as formulas II-IV; that is, too tacky in the freezer and loss of flexibility in the dessicator. All the formulas were totally deteriorated in an oven environment and exhibited varying changes in the refrigerator and at room conditions. Of all the formulas, formula VII, i.e., the formula with the least amount of glycerin, was most satisfactory on the day of manufacture as well as after storage at the various conditions excepting the oven. For this reason, it was chosen as the most suitable formula for further evaluation.

The preliminary work indicated that laminated aluminum foil bags appeared to be a suitable package, that temperatures above room temperature caused physical instability of the packaged foam, and that a loss of flexibility was not always associated with losses in moisture content. Also, preliminary studies indicated that proper storage conditions must be defined in terms of relative humidity as well as temperature.

As discussed earlier, a stable product must maintain potency and
utility after it has been packaged and stored for prolonged periods. To determine the stability of the foam as far as shelf-life and marketing potential were concerned, a three month storage study was conducted with stability evaluated at one month intervals according to physical properties, moisture content changes, and potency as determined by the semiquantitative microbiological assay.

On the day of manufacture, nitrofurazone foam was very flexible, not sticky, and dissolved on moist skin within fifteen seconds; these properties make it a very useful dressing for traumatized and infected skin. On the day of manufacture, moisture content was 9.7%. After one, two, and three months' storage in a refrigerator, freezer, and at room conditions, the moisture content varied insignificantly ($P > 0.3$) and the foam retained its original physical properties. However, in a dessicator, the moisture content decreased significantly ($P < 0.05 > 0.001$) and the foam lost flexibility. The data in Table XIV show that the package is a suitable container for storage at high humidities as it prevented the foams from gaining excess moisture which would have caused them to become too tacky and difficult to handle. At low humidities, however, the package did not prevent the foam from losing moisture. Preliminary work showed some formulas will lose flexibility without a corresponding decrease in moisture content, suggesting that some unknown inherent factors were causing the foam to lose flexibility. However, in this three month study, the loss of flexibility was associated with a significant loss of moisture; this indicates that the modified formula, i.e., VII, does not in itself contribute to loss of flexibility. Why the moisture impermeable foil bags were not effective in preventing loss of moisture is elusory. Speculation would lead one to believe that
either the seals were ineffective or that the foil bags are not really impermeable. Because the sealing procedure was shown to be effective and samples stored at high humidities did not gain moisture, these speculative reasons can probably be eliminated. In general, the only conclusion one can make from these data is that a relative humidity of 5-6% will cause nitrofurazone dry foam to lose moisture and flexibility even when packaged in foil laminates.

To determine if the storage conditions, time, and/or the package caused any deterioration of the drug in the foam or to determine if the vehicle itself inactivated the drug, a microbiological assay was done. A product must retain at least 90% of the labelled potency after storage for prolonged periods if it is to be accepted as therapeutically efficacious. Table XV shows the zones of inhibition both for the nitrofurazone foam and the commercially available nitrofurazone cream obtained on the day of manufacture and after one, two, and three months' storage at the four storage conditions. Storage of the nitrofurazone dry foam at room conditions, freezer, and a dessicator for one and two months did not produce statistically different diameters of zones from those produced on the day of manufacture. Samples stored in a refrigerator for one and two months, however, did produce statistically different zones as did storage for three months at all four conditions. These statistically different zones after storage were all greater than those on the day of manufacture. To explain these differences is difficult although two possibilities exist. One is that refrigeration conditions or long storage causes some interaction to occur between nitrofurazone and one of the components of the vehicle (e.g., Miranol) such that a greater release occurs. What this interaction might be cannot be
ascertained at this time. However, when comparing differences between zones after one and two months with zones after three months, there is no significant difference and this fact casts a doubt on the possibility of an interaction resulting in increased release. The second explanation involves the manufacturing process. All the samples were from the same batch, but not necessarily from the same pans. Because some pans became slightly warped from use, a difference in the thickness of the foam between pans and even within the same pan may have occurred; also, differences in bubble size may have occurred since it was not very easy to obtain a consistent and uniform spreading procedure. Thus, samples which were thicker or had small bubbles would have contained more active ingredient and produced larger zones; similarly, thinner samples or samples with larger bubbles would produce smaller zones. This second possibility seems much more likely to explain the statistical differences observed with the nitrofurazone dry foam.

On the day of manufacture, silver sulfadiazine foam was also very flexible, not sticky, and dissolved on moist skin in fifteen seconds. Table XVI summarizes the moisture contents on the day of manufacture and after one, two, and three months' storage at the four storage conditions. Silver sulfadiazine dry foam packaged in laminated aluminum foil bags and stored for three months in a freezer, refrigerator, and at room conditions showed similarities with nitrofurazone foam; that is, physical properties and moisture content demonstrated statistically insignificant changes after one, two, and three months' storage. Storage in a dessicator produced surprisingly interesting results. It would be expected from the nitrofurazone results that the silver sulfadiazine stored in a dessicator would also lose moisture and
flexibility. However, this was not the case at all; after one, two, and three months' storage, moisture content was statistically unchanged and flexibility was maintained. Because identical procedures with respect to manufacturing, packaging, storage, and evaluation were employed for both the nitrofurazone and silver sulfadiazine dry foams, the different results can only be attributed to the active ingredients—namely that silver sulfadiazine somehow aids in retaining moisture and flexibility while nitrofurazone does not. Other active ingredients may very well exhibit similar types of effects and this must be considered during any future drug evaluations.

As with nitrofurazone, potency as a guide to stability of silver sulfadiazine dry foam, was determined by comparing the zones of inhibition on the day of manufacture and after one, two, and three months' storage. Table XVII shows the zones for both the foam and the commercially available cream. Samples stored for one and two months at all four conditions produced zones of inhibition which were statistically insignificant from the zones obtained on the day of manufacture. Three months' storage in a refrigerator, freezer, and dessicator also did not affect the potency of the dry foam while samples stored at room conditions for three months produced statistically smaller zones \( (P < .001) \) when compared to the day of manufacture. While the possibility that three months' storage at room conditions could have caused a loss of potency, this difference was more likely due to variations in sample thickness and/or bubble size as discussed earlier.

For use on open, infected lesions, use of a sterile product is desirable. Sterilization of the dry foam could easily be achieved by three of the four methods employed. Aseptic methods produced a sterile
product substantiated by no growth in the two accepted test media, fluid thioglycollate medium and soybean casein digest medium. Growth occurred in the positive controls which indicated that the media and incubation conditions were conducive to microbial growth. Because aseptic preparation requires strict adherence to technique procedures and is always subject to accidental contamination, even if good technique is employed, sterilization of the final, packaged product is more advantageous from a manufacturer's viewpoint.

Ethylene oxide is widely used to sterilize products which cannot be autoclaved and which can be packaged in ethylene oxide permeable containers, namely plastics. Even though this product must be packaged in foil, it could be packaged first in plastic, exposed to ethylene oxide, and then packaged as a double bag with a laminated aluminum foil bag as the outer container. For this reason, contaminated foam was packaged in plastic and sterilized in a cold ethylene oxide cycle, i.e., 29° for three hours, followed by aeration for eight hours at 63°. Upon removal of the foam, it was noted that all flexibility was lost; the product was friable and crumbled easily. It was felt that this was due to the temperature employed in the aeration cycle and not due to a direct ethylene oxide chemical reaction. Even though sterility was achieved as shown by no growth in the testing media, the deteriorating effects of the aeration cycle excludes ethylene oxide as a method of sterilizing this product.

Method IV, gamma irradiation, is the method of choice for products packaged in laminated aluminum foil. Packaged samples which were exposed to radiation conditions were indeed rendered sterile as no growth was noted in the testing media; just as important was the fact that the foam
retained adequate flexibility and utility. Thus, this method provides a very practical, inexpensive, and efficient means to sterilize the packaged dry foam. It should be mentioned that all the methods of sterilization were performed on unmedicated samples. Thus, radiation will not be a useful method if the drug or drugs incorporated into the foam are destroyed or undesirably altered by radiation.

In order to determine if the foam could withstand normal shipping and mailing procedures, packaged samples were shipped by mail to the East Coast and returned unopened to the laboratory. The same packages were sent immediately to a second West Coast city and again returned unopened. These samples which were exposed to common mail handling were opened and compared to samples kept in the laboratory. No adverse changes were noted in the shipped samples as they all retained their original flexibility and in general, showed that the product could very satisfactorily withstand any conditions of shipping and mailing.
A highly water soluble, flexible dry foam was recently developed (1-3). The dry foam dissolves quickly on moist skin and releases the active ingredient without inunction. However, the foam is sensitive to moisture such that a loss of moisture causes the foam to lose flexibility while a gain causes it to adhere to the package or liquefy when touched.

The objectives of this study were to 1) modify the formula to minimize moisture sensitivity, 2) determine a suitable package, 3) determine proper storage conditions, 4) determine shelf-life of nitrofurazone and silver sulfadiazine dry foams, and 5) determine a suitable sterilizing procedure since the dry foam is intended for use on open, infected lesions.

Six different modified formulas were made and compared to the original. Glycerin, in varying concentrations, was used as the modifying agent since its humectant properties would help retain moisture and hence, flexibility. Comparison of the six formulas showed a glycerin concentration of 0.19% wet weight produced the most satisfactory dry foam with respect to flexibility, nonstickiness, dissolution time, and porosity. Using this modified formula, the remaining objectives were accomplished.

Plastic bags and plastic with liners proved unsatisfactory as a packaging material as they were permeable to moisture. On the other hand, samples of dry foam packaged in laminated aluminum foil bags and
stored at room conditions, in a refrigerator, and in a freezer, did not gain or lose moisture. However, aluminum laminates did not prevent loss of moisture and flexibility in samples stored at elevated temperature, i.e., 37°, or low humidities, i.e., 5-6%. Because the latter storage conditions are impractical, the unsatisfactory results are only academic. Consequently, laminated aluminum foil bags provide a suitable packaging material for the dry foam.

While storage at room conditions and in a freezer indicated the dry foam was physically stable for at least three months, refrigeration conditions appeared to be most satisfactory. Both temperature and relative humidity will physically alter the dry foam and these parameters can best be controlled in a refrigeration environment. Thus, 5° with a relative humidity of 55-60%, the usual refrigeration conditions, are recommended for storage of the dry foam.

The study also indicated that the foam can physically withstand common shipping and mailing procedures; thus, mailing of the final product does not pose any problems.

Potency of nitrofurazone and silver sulfadiazine dry foams was not significantly affected by any component of the vehicle, by the package, or by any of the storage conditions including the two extremes of relative humidity, 75-90% and 5-6%. Thus, at least these two drugs can be incorporated successfully into the dry foam. This suggests that the dry foam might be a satisfactory vehicle for other commonly used antimicrobials.

Sterilization with ethylene oxide causes the foam to lose flexibility and thus, this method cannot be used. While aseptic preparation employing autoclaving or filtration of the casting solution produced
a sterile product, the best method, with respect to expense, practicality, and efficiency, was sterilization by gamma irradiation. This method allows the final packaged product to be sterilized without physical degradation. Gamma irradiation is therefore recommended as the sterilization procedure provided the active ingredients are not undesirably affected by radiation.
BIBLIOGRAPHY


