# BACTERIOLOGICAL AND DERMATOLOGICAL TESTING OF COSMETICS\*

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TESTING THE multiple characteristics and properties of cosmetics involves a great number of methods. This is due to the various preparations of cosmetics. For this reason, a complete investigation of a cosmetic consists of determining (1) its harmlessness; (2) its purity; (3) the value of its preservative agents; (4) its germicidal or fungicidal value, if any; and (5) its performance, and substantiating the claims made by its manufacturer.

# (1) Tests for Harmless ness

The tests for harmlessness can be divided into: toxicity tests and skin irritation tests. Toxicity tests indicate whether or not a cosmetic is toxic and consequently dangerous when absorbed normally or accidentally by ingestion, when injected in the blood or when absorbed through the skin or mucous membranes. These tests are conducted by injecting the products of their extracts either intraperitoneally or intravenously, or by feeding animals a diet in which 10 or 20 per cent has

\* Presented at the Dec. 3, 1947, Meeting, New York City.

been replaced by the cosmetic. It was not rare a few years ago to find cosmetics that were toxic enough to cause death within a few hours. Today this happens very seldom although new cosmetics have been found recently to cause death within less than three days. A toxicity test is not complete if the determination of the minimum lethal dose has not been made. By injecting intraperitoneally various amounts of the product it is possible to determine the smallest dose that will cause death of the animals within three days. The safest method consists in obtaining the death of all animals with this dose; however, another method called minimum lethal dose 50 consists in seeking the death of only 50 per cent of the animals iniected.

The skin irritation tests should actually be called tissue irritation tests because they do not only involve the skin but also mucous membranes, the conjunctiva, and other tissues. All cosmetics submitted to us for skin irritation tests are first extracted and injected under aseptic conditions intradermally into ani-

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mals. Very small amounts are used and the results obtained, if positive, give a very good indication that the compound contains primary irritants. Comparison of the results of this test with short duration patch tests such as the United States Navy skin irritation test or the soap manufacturers' patch test have indicated that this method is just as reliable. It is also much easier to apply and the results are obtained in a much shorter time.

The eye irritation test consists in dropping a couple of millimeter cubes of the cosmetic on a rabbit's eye and then examine the degree of irritation so obtained. When these preliminary tests, intraperitoneal test or eye irritation tests, give a negative result, i.e., when no skin irritants are detectable we also conduct a patch test. Whenever possible we follow the method of Drs. Louis Schwartz and Samuel Peck which we have found to be the most suitable for the greatest number of cosmetics. It consists in applying the cosmetic on human skin for a period of five days. If the cosmetic is in solid form it is applied as is; if it is liquid it is tested by soaking a freshly boiled-off and sterilized piece of gauze in the solution to be tested. The patch is covered by means of water-proof surgical tape and removed at the end of a period of five days. The skin is then observed for the presence of skin irritation that very day and the following two days. Ten to fifteen days after the removal of the first patch another patch is applied for forty-eight hours after

which the skin is observed that day and the following two days. The presence of skin irritation caused by this second patch indicates -bat the cosmetic so tested is a cutaneous sensitizer. These tests are conducted first on 50 people. When five or more individuals show an irritation the test is considered as being completed and the cosmetic is reported as being a primary irritant. When two or less than two individuals show a reaction, the test is completed to a greater number of individuals never to be inferior to 200. These tests may probably look too severe but they are not. We have found a great many cosmetics that, when tested prior to marketing, were found to contain either primary irritants or cutaneous sensitizers, or both. It was definitely easier for the manufacturer to change his formula before launching the cosmetic than to market it without testing and face a great many costly lawsuits against which he would have had no defense.

## (2) TESTS FOR PURITY

Very often cosmetics which have irritating properties contain impurities. This is not always the case, but it is nevertheless advisable to have purity tests conducted on all types of cosmetics. In the spirit of the Food, Drugs and Cosmetic Act, the purity of a compound comes next to its harmlessness. The first step in conducting purity tests on cosmetics is to make bacterial and fungal counts. Every manufacturer will agree that if two cosmetics con-

tain very similar, not to say identical compounds, the one which will have the lowest bacterial and fungal counts will be of better grade and consequently have more merchandising value. We have found amazingly high counts in cosmetics, so high that some of them almost defy comparison with any other polluted source of bacterial or fungal life. On the other hand, a great many cosmetic chemists have during the past years endeavored to produce cosmetics that were of a great bacteriological purity and sometimes even germ and fungus free. This state of purity is verified by conducting a sterility test according to the method. described in the U. S. Pharmacopœia. According to the test method, a cosmetic is sterile after it is placed with aseptic precautions in a tube of sterile broth and kept at a temperature of 7° C. for seven days, the tube does not show any growth and remains crystal clear. Sterility tests are of course important when cosmetics may be absorbed in the digestive tract.

# (3) Tests for Preservatives

Some cosmetics pass the purity test described above immediately after they are manufactured and for a short period of time. Then, when their shelf life begins in department or drugstores, their bacterial and fungal count increases to amazing heights. In order to remedy this cause of deterioration, cosmetic chemists add preservatives to their formula with the result that the shelf life of their products is greatly increased. It is always very wise to test the value of the preservatives contained in these cosmetics. This is generally done by measuring the bacteriostatic and fungistatic properties of the cosmetic itself. Very often we are asked to first test the compound which the chemist plans to use in order to determine the concentration at which it should be added to the formula. Bacteriostatic and fungistatic tests are very simple and rapid and are conducted according to the method of the Federal Department of Agriculture. These properties are verified by plating the sample of the product in Petri dishes planted with bacteria or fungi and measuring the width of the zone of inhibition where the bacteria or fungi do not grow on the plates. A cosmetic is said to be bacteriostatic when it inhibits the growth of bacteria, and fungistatic when it inhibits the growth of fungi. Needless to say, these preservatives should be tested for toxicity, skin irritants, and cutaneous sensitizers at the concentration at which they are to be used in the cosmetic prior to being incorporated in the formula. We have tested more than 375 germicides-fungicides capable of being used as preservatives in cosmetics because of their bacteriostatic and fungistatic properties. When each were patch-tested at the concentration where they did meet the requirements of the cosmetic chemists, they were found to be highly irritating, or sensitizing, to such an extent that out of this huge number of various compounds less than a

half dozen were found to be suitable for use in cosmetics.

# (4) Tests for Germicidal or Fungicidal Value

Cosmetics can be so treated as to be not only bacteriostatic or fungistatic, but also germicidal or fungicidal, which means that they not only inhibit the growth of bacteria or fungi but actually kill them by contact. The germicidal or fungicidal value can be determined by making a phenol coefficient determination or by using Barail's contamination test. The safest way to determine the germicidal or fungicidal value of cosmetics is to use our contamination method which has been published several years ago and adopted by many bacteriological laboratories. It can be used by any bacteriologist and gives results in forty-eight hours. The compounds mentioned before as being bacteriostatic and fungistatic agents suitable for cosmetics are also used to give germicidal and fungicidal properties to various kinds of cosmetics.

# Selection of Tests

From the description of the various tests it is obvious that most of them are applicable to all types of cosmetics. Except for the eye injection and the feeding toxicity test it can be said that all other tests can be applied to all cosmetics.

We can divide cosmetics into three groups according to the nature of the tests to which they should be submitted prior to marketing.

The first group includes mascara,

eyelid kohl, eye lotions, face powders, face lotions, soaps, shampoos, detergents, hair lacquers, hair dyes, hair bleaches, hair tonics, hair rinses, and cold wave lotions. Thev are tested for toxicity by intraperitoneal injection and their minimum lethal dose should be determined. Their irritating and sensitizing properties are evaluated by means of intradermal injections and patch tests on at least 200 people. They are of course tested for purity. As all these cosmetics may actually come in contact with the eve, eve injection tests should also be performed by all means on all of these compounds. This is obvious when many eye lotions and eye creams are concerned but there have been serious accidents caused by each and every one of the other types, such as when a single drop of shampoo, hair bleach, cold wave lotion, or any other one accidentally falls in the eye.

The second group includes toothpastes, dentifrices of all kinds, mouth washes, lipsticks, nail polishes, and cuticle softeners. They are tested for toxicity by intraperitoneal injection, determination of the minimum lethal dose, and an animal injection and patch tests should determine whether they are irritating and sensitizing, or not. Tests for purity are conducted and also feeding tests. Everybody will understand that feeding tests have to be conducted on toothpastes, mouth washes, and lipsticks. Cases of toxicity due to nail polishes and cuticle softeners have been reported

by nail biting individuals or people who just suck their fingers: not only children but grownups too.

The third group includes face creams, cake make-up, shaving creams, after-shave lotions, hemostatic cakes, rouge, suntan lotions, deodorants, anti-perspirants, hand lotions, foot powders, leg make-up, colognes, and perfumes. It does not appear necessary to conduct eye injection tests or feeding tests on cosmetics of this group although we have seen cases where it was necessarv to do so because of accidents that had caused serious lesions. This group is tested for toxicity by intraperitoneal injection and minimum lethal dose and for primary irritants and cutaneous sensitizers by animal injection and patch tests. Purity tests are conducted as on the other two groups as explained. Furthermore, tests for preservatives, bacteriostatic and fungistatic propgermicidal or fungicidal erties. values are eventually conducted on each of these three groups of cosmetics if the tested sample is supposed to contain preservatives or to have been treated in order to render it germicidal and fungicidal.

## (5) Performance Tests

The cosmetics of the *first group* will be tested as follows for performance and efficiency. Mascara should be free from clots, adhere to the lashes, and not be washed off by water (rain) or tears. Eyelid kohl should not smear or spread: its pigment should be distributed evenly and the finished product should not

be water soluble. These properties of mascara and kohl can be verified in the laboratory. The claims made by the manufacturers of eye lotions can be verified only in field tests on large consumers panels. Face powders can be tested in the laboratory for particle size and their adhesive properties in regard to skin; however, the tests should be conducted on large consumers panels. Face lotions can be tested in laborafor tories cleansing properties claims concerning imbut all provement of skin conditions should be verified in the field. Soap shampoos and detergents can be tested in vitro for cleanliness, formation of suds, appearance of the hair, formation of a film, but the most conclusive results will be obtained in field tests involving a great number of people of all ages, of both sexes and of various colors and textures of hair. This of course, applies to hair lacquers, hair dyes, hair tonics, and cold wave lotions which cannot be efficiently tested in the laboratory. Among the cosmetics of group 2, toothpastes, dentifrices, and mouth washes can be tested in vitro for efficiency. On the other hand, consumers panels are indicated when comparative tests are involved. Lipsticks and cuticle softeners can only be tested in the field. Some cosmetics of group 3 can be tested only in the field: such are face creams, cake make-up, shaving creams, after-shave lotions, rouge, hand lotions, leg make-up, colognes, and perfumes. Suntan lotions can be first tested in the laboratory in

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order to determine whether they actually form a film which is impervious to ultra-violet light. Deodorants are first tested in vitro and their efficiency measured by means of our osmometer, which is the only scientific apparatus giving in one single operation the odor threshold number of any compound. Testing of deodorants in the laboratory is always made with our artificial perspiration whose formula is identical to that of human perspiration and which is always kept fresh and with proper bacterial insemination. Finally foot powders can be studied for their fungistatic properties, against athlete's foot organisms and for deodorizing properties by means of our osmometer. Of course, the best results on suntan lotions, deodorants, and foot powders in the field will be obtained only after these preliminary tests will have been conducted in the laboratory and given satisfactory results.

#### Conclusions

As that of a physician, the first preoccupation and worry of a cosmetic chemist should be and most of the time is: *primum non nocere* (not to cause any harm). His second worry is, of course, to combine experience, technology, and cleanliness in order to obtain the cleanest and most appealing products.

At this stage, he is very close to his goal, the production of a highclass product of great efficiency and innocuity.

In testing his final product, or its component parts, the cosmetic bacteriologist and the cosmetic dermatologist will give the cosmetic chemist the best service and advice for many reasons. First of all some cosmetic bacteriologists and cosmetic dermatologists are also cosmetic chemists. Those who are not. know enough cosmetology to understand the problems they are confronted with and to satisfactorily solve them. Finally, after many years of experience, all problems which appear complicated to the beginner or the layman, are just repeat performances to the seasoned cosmetic bacteriologist or cosmetic dermatologist, and both can handle them in the shortest possible time and with the greatest chance of success. Second, everyone will agree that some of these tests require either a specialized staff and equipment, or an organization with outside contacts and ramifications that very few cosmetic chemists have.

Testing cosmetics in accordance with the requirements of the Food, Drug, and Cosmetic Act and the Federal Trade Commission and in compliance with the desiderata, hopes, and wishes of cosmetic chemists is not always easy. Yet it is never boresome; sometimes it is fun. Personally, we find it always gratifying because we always endeavor to account for your hard work and sleepless nights and enjoy giving you the final OK that means so much to you.