THE ROLE OF GLYCEROL IN SKIN HYDRATION

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The utility of glycerol in cosmetic formulations is well established. Thus, glycerol has been used as a humectant, lubricant, plasticizer, binder, spreading agent, etc.

For many years, one of the functions regularly attributed to glycerol was a "softening" action on skin. That glycerol *per se* does not have a softening effect on keratin has been shown clearly by investigators during the past ten years (1–3). Indeed in aqueous products containing glycerol, the glycerol may actually serve to slow down the hydration of keratin when these products are used on the skin. This may be illustrated by soaking strips of desiccated callus in various glycerol-water solutions and studying their hydration by observing the rate at which they swell, soften, and become opaque (hydrated callus is opaque, desiccated callus is translucent). Results indicate that callus hydrates most rapidly in pure water, and the addition of 5 per cent glycerol has an appreciable effect of slowing down the rate of hydration. Higher concentrations of glycerol slow down the hydration even more markedly.

Flesch (4) recently reported the results of experiments in which the rate of moisture loss from skin was studied. This was accomplished by mounting a piece of excised skin over the mouth of a vessel filled with water and then measuring moisture loss through this piece of skin by daily weighings. When a film of glycerol is placed on this skin membrane, the rate of moisture loss from that system is decreased. From these results, the author concluded that glycerol decreased the diffusion of water through the skin membrane probably *via* occlusion of the skin surface. An alternate explanation of these results can be postulated, however, that does not involve any alteration of the diffusion rate of water through the skin membrane. Thus some of the moisture lost through the skin membrane could readily have been trapped by the glycerol film, thereby lowering the rate at which moisture was being lost from the complete system. Indeed this would be expected, since glycerol is more hygroscopic than skin. This would mean that the skin would lose moisture at the same rate with and without a surface film of glycerol, but, in the case of glycerol coating, some of this moisture would not escape from the system.

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Powers and Fox (5) studied the effect of various cosmetic ingredients (including glycerol) on the rate of moisture loss from the skin in an *in vivo* situation. In their experiments, small desiccator cups containing silica gel were strapped on the skin, and the rate of water loss measured from an untreated site in comparison to a site rubbed with anhydrous glycerol. The authors found that glycerol increased the rate at which water was lost from the skin. They conclude that glycerol accelerates the diffusion of water from moist dermal tissue to the surface, and that this accounts for the beneficial effect of humectants on skin during dry weather (6).

It has been shown that the rate at which moisture diffuses to the surface of the skin is governed by a rate regulating barrier membrane (1). If Powers and Fox's interpretation of their data is correct, one must conclude that glycerol increases the rate of water diffusion through this barrier membrane. This does not appear to be a likely action of glycerol.

Another possible explanation for these results is that during the process of rubbing the anhydrous glycerol on the skin, it picked up water from the air and the weight changes really measured water lost by the skin plus water lost by the glycerol.

An experiment was set up to determine how much water could be taken up from the air by 4.9 cm.^{2} surface area of glycerol (equivalent to the surface area of the desiccator cups used by Powers and Fox) in short periods of time. Exposing 4.9 cm.^{2} of anhydrous glycerol to 65 per cent R.H. for one minute resulted in a moisture uptake of 3.8 mg., 3.0 mg. and 4.5 mg. on successive trials.

In a second experiment, about 30 mg. of anhydrous glycerol was spread over an area of 4.9 cm.^{2} using a glass stirring rod in a manner analogous to that described by the authors. (This amount of glycerol corresponds to the amount of glycerol Powers and Fox had covering their one-inch circular area.) After spreading the glycerol for one minute at 65 per cent R.H., the moisture uptake was determined. The results were 3.9 mg. and 3.5 mg. on successive trials.

In both experiments, short, wide-mouthed, glass weighing bottles were used in which a circular area of 4.9 cm.^{2} was inscribed on the bottom. In the first experiment, glycerol was dropped into the vessel until it formed a circular drop equal to the inscribed area. In the second experiment, 30 mg. of glycerol was placed in the center of the inscribed area and spread with a micro glass stirring rod.

These values are of the right magnitude to explain the increased moisture loss observed by Powers and Fox from the glycerol treated site. Thus, they report increased moisture loss on one subject of 3.6 mg., 4.4 mg. and 3.6 mg. during three successive hourly measurements, and on a second subject of 1.7 mg., 1.5 mg., 1.4 mg. and 1.9 mg. during four successive hourly measurements. Since the desiccant cups were removed each hour

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for weighing, this probably gave the glycerol a chance to rehydrate by taking up water from the air.

Thus, it is indeed possible that the results obtained by Powers and Fox can be fully explained by water loss from the hydrated glycerol rather than an increase in water loss from the skin.

It has recently been reported by Flesch (4) that glycerol plus callus exhibits unusual humectant properties, over and above those which would be predicted from the humectancy of glycerol and callus taken separately. These results have led to the speculation that glycerol penetrates between the keratin fibers of the horny layer and facilitates the absorption of water.

Flesch studied the moisture uptake of glycerol, callus and callus-glycerol mixtures when exposed to 100 per cent R.H. Under such conditions, minor fluctuations in temperature can cause water to condense. Furthermore, the attainment of equilibrium conditions with glycerol is virtually impossible since glycerol is miscible with water in all proportions. In any case, the author's results actually show that the *rate* at which a mixture of glycerol and callus picks up moisture is greater than would be predicted from the rate at which callus alone and glycerol alone pick up moisture.

The fact that Flesch is dealing with rates rather than equilibrium conditions is most apparent when one considers that his data show that at the conclusion of this experiment, glycerol had taken up about 130 per cent of its own weight in moisture. At a R.H. in the high 90's, one would expect glycerol to take up well over 900 per cent of its own weight in water (5).

When dealing with rates of water uptake, such things as surface area of glycerol and callus, and size and shape of the vessel in which the glycerol and callus are placed become critical. One can readily see how a potentiation in rate of water uptake could be achieved in the glycerol callus system if the surface area of the glycerol were increased due to the presence of callus. With this in mind, an experiment was set up to test the hypothesis that the *rate* of water uptake of a glycerol-callus system was elevated above that which would be predicted by callus and glycerol alone, but that the *equilibrium* uptake was merely a summation of the water taken up by glycerol plus that taken up by the callus.

Into three glass vials, 50 mg. of ground callus (80 mesh) was weighed. To one of these vials was added 50 mg. of anhydrous glycerol and to a second 250 mg. of anhydrous glycerol was added. Into two other identical vials, about 50 mg. and 250 mg. of anhydrous glycerol was weighed. All five vials were then placed into a constant humidity chamber at 97 per cent R.H. and weighed at regular intervals. From the amounts of water taken up by the 50 mg. sample of callus alone and the 50 mg. and 250 mg. samples of glycerol alone, calculations were made as to how much water should be taken up by the two glycerol-callus mixtures, assuming a strictly addi-

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tive effect. These calculated values were then compared to the actually measured values for the two glycerol-callus systems (Results—Table 1).

TABLE 1—MOISTURE UPTAKE BY GLYCEROL-CALLUS SYSTEM

Days	50 mg. callus & 50 mg. glycerol Calculated	Observed	50 mg. callus & 250 mg. glycerol Calculated	Observed
1	39 mg. water	33 mg. water	82 mg. water	73 mg. water
2	50 mg. water	47 mg. water	122 mg. water	114 mg. water
4	68 mg. water	65 mg. water	184 mg. water	173 mg. water
7	84 mg. water	81 mg. water	247 mg. water	237 mg. water
11	104 mg. water	100 mg. water	315 mg. water	303 mg. water
14	114 mg. water	111 mg. water	354 mg. water	342 mg. water
18	115 (?) mg.	124 mg. water	396 mg. water	388 mg. water
21	130 mg. water	131 mg. water	429 mg. water	419 mg. water
25	141 mg. water	145 mg. water	468 mg. water	464 mg. water
28	147 mg. water	151 mg. water	492 mg. water	490 mg. water

As the results indicated, in neither case was a "potentiation" of water uptake seen. It should be noted that even after twenty-eight days, neither system had reached equilibrium. The reason for our inability to demonstrate the increased rate of water uptake reported by Flesch is not clear. It is most probably related to the fact that the size and shape of the vessels used by Flesch were better suited to increase the surface area of glycerol in the glycerol-callus system than were the vessels used in this experiment. In any case, it is clear that the glycerol-callus system does not possess any enhanced ability to bind moisture over and above that which would be predicted on the basis of water uptake by glycerol alone and callus alone.

Thus the functional role of glycerol on the skin, from a biological standpoint, has not clearly been established. Whether it has some specific biological action remains to be determined.

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NOTE: A recent publication by Fox, *et al.* [*J. Soc. Cosmetic Chemists*, **13**, 263 (1962)] has clearly shown that the enhanced water sorption of glycerol-callus systems, reported by Flesch, was a result of rate rather than equilibrium measurements.

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