SYMPOSIUM: TOXICOLOGY AND BIOCHEMISTRY OF FOOD ADDITIVES USED IN FATS AND OILS

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Safety of Emulsifiers in Fats and Oils[1]

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ABSTRACT

Consumers have come to expect many foods that can be served with minimum effort. Such convenience foods frequently require the use of emulsifiers in their formulations. Emulsifiers enable fatty and aqueous phases to be combined. Various emulsifiers have been developed to meet specific needs and have been judged adequately safe for their intended uses. Safety evaluation programs may include both feeding studies and metabolic studies. Feeding studies establish the levels at which a compound can be fed to experimental animals with no detectable ill effects. From this information, it is possible to estimate quantities that may be consumed safely by humans. Metabolic studies tell how a compound is handled by the body—whether it is burned for energy, stored, or excreted. The choice of studies to use in evaluating a specific compound depends upon the chemical nature of the compound, its similarity to familiar materials, and its intended use. Studies carried out with polysorbates and with monoglycerides are reviewed to illustrate these points and to show how safety testing programs yield the information needed for making sound safety judgments.

INTRODUCTION

Emulsifiers are among the most important of the food additives in terms of both volume and function. Their functional importance derives from their ability to make oil and water mix in the form of an emulsion. Emulsions are very common in the culinary art. Among the most familiar are milk, butter, mayonnaise, sauces, cake batter, and frostings. The emulsifier inherently present or first used in these foods was a complex mixture of phospholipid and protein.

Cake batter was one of the first foods to benefit from the use of man-made emulsifiers. Around 1930, it was found that shortening that contained some mono- and diglycerides could make a better cake than ordinary fats, with a greatly reduced incidence of cake failure (1-6). Since then, it has been found again and again that the use of man-made emulsifiers instead of the ones nature happened to provide, or in addition to them, makes it possible to prepare food products having greatly improved properties. In some cases, the new emulsifiers make it possible to prepare new, different foods that simply could not exist without the emulsifiers.

Consumers like and want the foods that are made possible by emulsifiers. When they respond to consumer surveys and when they lay down their money in the grocery store, they make it clear that they are sensitive to the benefits conferred by the emulsifiers.

Table I lists the emulsifiers that may be added to fats at the manufactuer's level (7). Several emulsifers that commonly are used in foods are not on this list, because they are not added to fats and oils as such. Most of the emulsifiers on this list are not used in fats sold for household use, but, rather, they may be used in fats sold for manufacturing purposes, such as to commercial bakeries.

All of these emulsifiers have been approved by the Food and Drug Administration for use in foods. Two of them are generally recognized as safe (GRAS), and the others have been approved under food additive regulations. Owing to limitations of time and space, details of the safety evaluation for each of these materials will not be recited. Only two of them are covered in detail below, but all of

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TABLE I

Emulsifiers Used in Fats and Oils

Mono- and diglycerides[a]	Propylene glycol esters
Polysorbates	Citric acid esters
Lactic acid esters	Ethoxylated monoglycerides
Polyglycerol esters	Sorbitan fatty acid esters
Tartaric acid esters[a]	Succinic acid esters

[a]Generally recognized as safe

them have been studied; and enough information has been developed about each of them to convince technically competent authorities that they are safe.

The two that will be covered in detail are the mono- and diglycerides and the polysorbates. There are two reasons for emphasizing them. First, they are more widely used than most of the other emulsifiers on the list. Second, their proofs of safety developed along different lines and can be used to illustrate the two different kinds of information that may go into safety evaluation studies. These are, first, feeding studies and, second, metabolic studies. In a feeding study, the material is fed to test animals to see whether it causes any harmful results. In a metabolic study, the material is fed to animals, and it and its biotransformation products are traced through the animal's system so that its metabolic fate can be determined. The two methods are complementary, and many safety evaluation programs use both; but usually one is more appropriate than the other for any given compound.

MONO- AND DIGLYCERIDES

The chemistry of mono-, di-, and triglycerides was worked out many years ago, and it was long theorized that triglycerides were broken down to mono- and diglycerides in the body during the processes of fat digestion. During the last 30 years, it has been demonstrated repeatedly that this is what actually happens. Enzymes, mostly pancreatic enzymes, hydrolyze triglyceride to monoglyceride and free fatty acid (8,9). These are absorbed into the intestinal wall and are used there to resynthesize triglycerides, which are transported on into the body through the lymphatic system (10-16). The monoglyceride that passes through the intestinal wall may be formed in the digestive tract by hydrolysis of triglyceride, or it may have been introduced into the digestive tract as such; it makes no difference, for the same compound is involved in either case.

In one set of experiments, rats were given either triglyceride or mixtures of triglycerides with mono- and diglyceride. Three hr later the material in their digestive tracts was analyzed. The same proportions of 1-monoglyceride, 2-monoglyceride, diglyceride, triglyceride, and free fatty acid were found, regardless of which material was fed, except when very high dosage levels of 2-monoglyceride temporarily had overwhelmed the capacity of the system to interconvert the different materials (17).

Nutrition experiments showed that monoglycerides serve very efficiently as energy sources. They are just as absorbable as triglycerides, and they provide just as much energy for growth, after a slight correction is made for the fact that partial glycerides contain a larger proportion of oxygen than triglycerides do (18,19).

A key point in the safety assessment of the partial glycerides is the conviction that it must be safe to consume small, incremental amounts of materials that are formed in the body naturally, continuously, and abundantly. This seems very reassuring, but a few more questions can be raised. One of these has to do with the possibility that monoglyceride in foods will influence the digestive process by lowering the surface tension in the digestive system. This was tested by Dasher (20), who showed that, although monoglyceride added to a cottonseed oil/buffer system does, indeed, lower the interfacial tension, this effect becomes insignificant when the interfacial tension already has been reduced greatly by the bile salts that are normally present in the digestive tract. Later work showed that monoglycerides help solubilize triglycerides into a solution of bile acid conjugates and thus aid in the digestion of fat (21).

The other questions that can be asked are vague ones, but no less important. What if feeding monoglycerides causes some effect that would not be anticipated from all that we know about their chemical and biological properties? There is really only one way to answer these questions, and that is by doing a feeding test, to see what effects the material really does have upon living systems. A number of feeding studies have been carried out with mono- and diglycerides dating back as far as 1941 (22).

Several mono- and diglycerides were fed to rats for 10 weeks, as 15% or 25% of the diet. Growth and feed efficiencies varied with the fatty acid compositions of the glycerides but not with glyceride type. The composition of depot fat laid down by the animals reflected fatty acid composition but not glyceride type. Necropsy and histology revealed no adverse effects that could be attributed to the partial glycerides (18).

In another experiment, 3 successive generations of rats were given diets that contained either 15% or 25% monoglyceride as the sole dietary fat. Growth, reproduction, lactation, and lipid absorption were no different from control animals fed cottonseed oil (23).

In still another study, groups of 64 weanling rats were fed for 11 months with rations containing 15% of partial glycerides from either lard or partially hydrogenated lard. Part of the animals then were sacrificed for histology, and part were used to initiate a 3 generation reproduction study. Growth, urinalyses, blood analyses, bone calcification studies, organ/body wt ratios, and reproductive performance were studied. The partial glyceride did not appear to be deleterious to the animals in any way (H.C. Hodge, unpublished results).

Later, a more elaborate study used groups of 100 rats, and they were kept on test for 2 years. The purpose of such a long term feeding study is to look for chronic effects of the test material, particularly for any possible increase in tumors, but no effects were found that could be attributed to the mono- and diglycerides (R.S. Harris and H.C. Hodge, unpublished results).

The few adverse effects that have been reported when high concentrations of partial glycerides were fed to animals have been associated with high levels of glycerides that contained only the saturated fatty acids. These effects, which are related to poor absorption of saturated fatty acids, were much the same effects as those seen whenever high concentrations of any saturated lipid are fed (22).

The final consideration in evaluating the safety of partial glycerides has to do with the quantities that are consumed. Special purpose shortenings sometimes contain up to 5% of partial glycerides, but the level in most shortenings is considerably less; and much of the fat in a normal diet contains only traces of them It is estimated that a normal daily diet contains, on the average, ca. 100 g total fat, which contains, in turn, ca. 1/2 g each of monoglyceride and diglyceride that has been added as emulsifiers. Probably another 1/2 g each of mono- and diglyceride comes into the diet from natural sources. The digestion of this dietary fat forms ca. 30 g monoglyceride in the digestive tract, so the amount that comes in as an intentional food additive is really of very little importance (24).

The entire subject of glyceride safety recently has been reviewed with very reassuring results (25).

POLYSORBATES

Whereas the structures of the mono- and diglycerides are familiar and reassuring, the structures of the polysorbate emulsifiers (Fig. 1) are unfamiliar, and the contemplation of them does not lead to an immediate assumption of their safety for food use. Therefore, the primary task in evaluating these materials has been to explore the effects that they produce in animals during feeding studies.

It initially was found that the polysorbates have very low acute oral toxicities. Doses of 30-40 g/kg gave only low incidences of death. The oral LD_{50} of the monooleate ester has been measured as 50 g/kg in rats and greater than 25 g/kg in mice (26-28). Thus, in an acute sense, the polysorbates are no more toxic than many other foodstuffs.

A 2 year feeding study in rats and a 3 generation reproduction study in rats both used a ration containing 2% of polysorbate 80, the oleate ester. In neither study was there any evidence of harmful results from this emulsifier (27,28). This kind of experience is a strong indication that a material will be suitable for human consumption.

The chronic toxicity of polysorbate 60 (the stearate ester) was studied by feeding rats for 2 years with rations containing 0, 2, 5, 10, and 25% of this emulsifier. All groups showed normal patterns of mortality. Males receiving the 25% level of emulsifier showed reduced wt gains after 12 weeks but not at later intervals, and all other groups grew normally throughout the study. Diarrhea was observed only among the rats receiving the two highest levels of emulsifier. The livers of animals receiving the 25% level showed some fatty changes of very slight degree, but no other microscopic changes were seen in any other tissues from any of the feeding levels (29).

The work of Chow, et al., (30) cast doubt on the meaningfulness of diarrhea as a clinical symptom in the rats that received high levels of polysorbate. Feeding purified casein diets that contained 5% polysorbate 60 to weanling rats produced diarrhea and growth retardation. But when the polysorbate was fed in a soybean meal diet, at either the 5% or the 15% level, there was neither diarrhea nor any other clinical manifestation, and histological examinations after 14 weeks showed no exceptional findings.

The most definitive long term study is the one described by Oser and Oser in 1956 (30-33). This study was a model of thoroughness for its time. Several of the polyoxyethylene sorbitan emulsifiers, individually and as a mixture, were fed to rats as 5, 10, or 20% of the diet for 2 years and through 4 generations. The rats were evaluated by many criteria, which can be summarized under the headings of growth, feed efficiency, clinical observations, reproductive efficiency, hematology, and histopathology.

The 5% level was chosen as representing a many-fold exaggeration over the level expected in human diets. The 20% level was chosen as one that should produce some harmful effects, and, indeed, some harmful effects were seen; most notably there was diarrhea in some of the groups at this extreme level. There were also some decreases in the survival of the new-born and some decreases in longevity. These effects were not seen at the lower levels, and the problems with diarrhea and reproduction could be alleviated by the addition of normal fat to the diet. These harmful effects would not be expected in a normal diet that contained both emulsifier and fat nor in any circumstances other than the grossly distorted conditions of this feeding study. The importance of feeding this high level of emulsifier was that it guided the researchers as to what they should look for at the lower levels.

It is most significant that not even the highest levels of emulsifiers gave any evidence of cumulative toxicity or of progressively changing physiological response through the 4 generations, and there was no evidence that the emulsifiers had any carcinogenic potential.

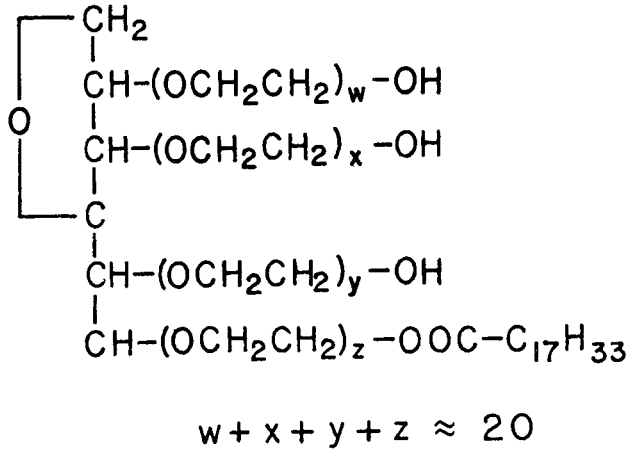

FIG. 1. Structure of polysorbate 80.

Other workers have fed polysorbate emulsifiers to mice, chicks, hamsters, dogs, and monkeys with no ill effects, except for the diarrhea that sometimes results from having high concentrations of polyoxyethylene sorbitan in the large intestine. These experiments have been summarized (34).

It has been suggested (35) that the polysorbate emulsifiers, like other surface-active agents, might influence the absorption of substances from the gastrointestinal tract. The use of surfactants to influence drug absorption is well known (36). The inclusion of polysorbate emulsifiers into diets at high levels has been reported to enhance the absorption of vitamin A but not the absorption of fat, while the effects on cholesterol absorption have been mixed (37). Eagle and Poling (26) observed marked hemosiderosis in hamsters receiving polyoxyethylene sorbitan monolaurate at levels of 5% or more in a purified diet and attributed it to enhanced absorption of iron. Wissler, et al., (38) established that the absorption of ^{59}Fe from the digestive tract of hamsters was increased when the hamsters were fed a ration containing polyoxyethylene sorbitan monolaurate. These animals showed elevated levels of iron in the blood, bone marrow, liver, large intestine, and cecum. It was speculated that the effect might be due to absorption of iron from the cecum when its contents became more fluid because of the presence of the emulsifier. There is no evidence that the polysorbates have any influence on intestinal absorption when they are consumed at levels corresponding to the levels at which they appear in practical human diets.

No matter how satisfactorilly a rat feeding experiment turns out, all it really can prove is that the material is safe for rats. There is necessarily a gap between the information derived from animal studies and the application of that information to human exposure situations. One of the chief ways to bridge that gap is to carry out metabolic studies that trace the fate of a compound through the biological system.

Experiments using radioisotope tracers showed that the ester link of the polysorbate molecule is split by intestinal lipase and that the fatty acid portion is released. This fatty acid becomes indistinguishable from the other fatty acids in the lipid pool of the organism, and it may be used to make lipids, or it may be burned for energy. The polyol moiety left after hydrolysis of the ester is very poorly absorbed from the intestinal tract, and most of it is passed out in the feces. Of the small amount that is absorbed, virtually all is excreted rapidly in the urine. This metabolic pattern has been worked out in detail in rats, using emulsifiers labeled with radioactive carbon in either the fatty acid or the polyol moiety (39). Experiments in humans have shown

that essentially the same pattern is followed (40). The emulsifier is split, most of the polyol is eliminated in the feces, and small amounts of polyol are excreted in the urine. Thus, man and rat metabolize the emulsifier in the same way and in a way that is recognized as innocuous. This finding lends strong support to the view that, since the material is safe for rats, it must also be safe for humans.

But to supplement this view further, there has been a number of human feeding studies published primarily because of interest in using polysorbates in emulsion-type diets for either experimental or therapeutic purposes. In most of these experiments, the subjects received relatively high levels of emulsifier for relatively short periods of time under close clinical supervision and showed no evidence of ill effects (27, 41-44).

All of the other emulsifiers that are approved for use in fats and oils have been subjected to a program of investigation, using some or all of the techniques mentioned above. Each of these programs has yielded data on which has been based a judgment that the material is adequately safe for its intended purpose.

SAFETY PHILOSOPHY

Almost all materials, whether naturally occurring or man-made, are toxic at some level of exposure, but the question of interest is whether they present a hazard at anticipated exposures. Thus, the data given above should be evaluated with reference to three basic philosophical considerations, which apply not only to the emulsifiers, but to all other chemicals in our environment, and even to the so-called natural foods that we have been eating for generations.

Under conditions of normal usage, these emulsifiers are not hazardous, but they cannot be said to be completely nontoxic. Some of them can cause harm to living organisms but only when they are misused by being fed at levels far greater than the levels to which people are exposed. Almost everything in our environment can be shown to be toxic, and we ought to be concerned, not with toxicity itself, but with harmfulness, which is a function of toxicity and exposure.

There is no way to prove anything, whether naturally occurring or man-made, to be absolutely safe. Consequently, these emulsifiers cannot be proved absolutely safe under all conditions. No matter how many experiments you run, no matter how many animals or humans you study, no matter how many criteria of well-being you consider, there is always the risk that some surprise may someday appear. Obviously, the more data we have, and the more reassuring those data are, then the lower the risk. However, the risk is never zero. This realization ought not to alarm us, for there are few things that any of us do that do not entail some level of risk, which we are willing to assume for the benefits that are derived at the same time.

The benefit/risk ratio must be evaluated. If a material offers tremendous benefits, then it is reasonable to use it despite a high level of risk. The traditional example is a life-saving drug whose function is valuable enough to justify using it despite a high incidence of dangerous side effects. The benefits conferred by the use of emulsifiers in foods obviously are nowhere near that great, but they are real; and, as mentioned above, it is known that they are perceived and appreciated by consumers. At the same time, the risk associated with their use is vanishingly small. This risk appears to be smaller than the risks associated with many of the natural foods that have been used for generations. (Many of the natural foods have not been studied the way these emulsifiers have been studied, so the risks associated with them are largely unknown.)

For these emulsifiers, which have been studied thoroughly, the benefits greatly outweigh the risks, and there is ample justification for continuing to use them to enhance the quality and variety of foods.

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