65.46 and 71.15 Å for the 1-butyro-3-palmitin and 1-butyro-3-stearin, respectively, can not at this time be associated with a given structure.

Summary

1. Six aceto- and butyroglycerides, including di- and triglycerides, were prepared and purified, and a number of their physical properties were determined.

2. Melting and transition points for each of the glycerides were determined by experiments with small portions of the samples in capillary tubes; the "thrust-in" technique was employed to detect some polymorphic transitions occurring in intervals as short as one second.

3. Using the data on melting and transition points, dilatometric data were obtained, insofar as possible, for the various crystalline modifications of the compounds as well as for the liquid state. From the dilatometric data calculations were made for expansibilities in the liquid and various solid states, and melting dilations for several solid states. In one instance the volume change on phase transition, without melting, was calculated.

4. From X-ray diffraction patterns the long and short crystal spacings for the highest melting forms of several of the glycerides were determined. On the basis of the short spacings, polymorphic designations commonly used for fats and oils were assigned. The long spacings obtained indicated a triple chain length structure for some of the compounds and as yet an undetermined structure for the other compounds.

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Letter to the Editor

March 12, 1956.

In the communication, "The Influence of Dietary Fat on the Glyceride Structure of Animal Fats," Reiser and Dieckert (1) state: "Kartha's concept is that all lipases have equal affinity for all fatty acids and *vice versa*" and "although Kartha has assumed that lipases are unselective in their action, such is certainly not the case." These statements make it appear that perhaps Kartha's views should be stated more clearly and completely. The explanation of glyceride structure of natural fats suggested by Kartha (2, 3) was based on the following evidence found in the literature: a) plant and animal lipases are non-specific towards the alpha and beta hydroxyls of the glycerol (4); b) plant lipases are inactive on acids of lower chain-length than C_7 and act non-specifically on all fatty acids above this (5); c) adipose tissue lipases (same properties as pancreas lipase) are inactive on acetic acid but with other saturated acids show increased rates of action as chain-length decreases, and with triglyceride of unsaturated acids from C_{16}–C_{22} reactivity at lower temperatures increases with the number of double bonds (6); d) all lipases act reversibly (7); e) in animal depots as also in maturing seeds, fats are always in presence of active lipase (8).

The reversibility of lipatic reaction leads to dynamic equilibrium, which eliminates the influence of specificities of action of lipases as regards the ultimate distribution of the acids among the glycerol molecules. In such a system there will naturally be continuous interchange of acid radicals between different positions of the same or different glycerol molecules and all initially formed products will be rearranged gradually in the direction of probability requirements wherein the final structure will depend entirely on the proportions of the different fatty acids. In such systems deviation from simple chance distribution is possible only on the basis of limits in the formation of some of the triglycerides required according to chance.

Reiser *et al.* (1) further quote Kartha as saying that "in adipose tissues, as well as mammary glands of animals, fat can be deposited from ingested foods without affecting the normal glyceride type of distribution." The conditions under which ingested fats can be deposited without disturbing the normal glyceride structure pattern have been described elsewhere (9); under other conditions distribution patterns can change within certain limits (9).

The evidence of Reiser *et al.* that endogenous animal fats may contain GS_3 in proportions measurably higher than those required according to random distribution (1) would, if confirmed, raise serious doubt of the validity of the restricted random distribution theory, at least when applied to animal fats. However the isotope dilution method used by them for the GS_3 determinations is based on the premise, which has yet to be established, that added labelled GS_3 does not accumulate in the precipitates or the mother liquors in the course of several crystallizations. If the isolated GS_3 contains less than the representative quantity of labelled tripalmitin, the result of the determination of GS_3 will be too high.

The component acids of the GS_3 of both chicken and rat fats are predominantly palmitic, but both fats contain significant and variable quantities of stearic acid (10). It is therefore possible that some of the values for GS_3 found by R and D are too high because the labelled tripalmitin in the recrystallized fractions was not representative of the whole of the GS_3.

Furthermore, because of the relatively high proportions of palmitic acid in the GS_3, the component acids of the other fractions being predominantly C_{18} or higher, the results reported by R and D in terms of weight percentage are misleading. Recalculation of the results in terms of mol percentage on the as-

sumption that all of the GS_3 is tripalmitin and all other triglyceride molecules are triolein gives values for GS_3 found/GS_3 calculated $\times$ 100 which are much lower than those recorded. For instance, in Table II this value for the fat from rats fed the basal diet is 107%. When recalculated in terms of mol percentage the value is reduced to 93%. Although the actual difference may be less than this, it is very likely significant.

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For A. R. S. KARTHA
by R. J. VANDER WAL

Letter to the Editor

Dr. Kartha has been kind enough to send me his response to Dr. Dieckert's and my paper on "The Influence of Dietary Fat on the Glyceride Structure of Animal Fats" and requested my comments.

We believe that the early part of his letter, with citations, confirms our statements, which he quotes, outlining his position on the lack of specificity of lipases. (The exceptional behavior of the lower fatty acids is not a factor since we were discussing animal depot fat.) Perhaps he meant to present these citations as further defense and elucidation of his position, rather than to correct our misrepresentation of it. His letter is not quite clear on this point.

One can find supporting evidence for almost any point of view by quotations from the literature, especially in this field. It was for that reason we felt that more experimental evidence, under controlled conditions, was required.

Kartha's criticism of our procedure can only be valid if it can be shown that the procedure is enough in error to affect the conclusions. He raises two objections.

a) Our isotope dilution procedure is faulty since we added tripalmitin as a carrier while natural saturated triglycerides contain some stearic acid.

Theoretically a carrier should be exactly the same substance as that being isolated. At the time we decided on the use of labeled tripalmitin as a carrier for total saturated triglycerides, the probable influence of the small amounts of tristearin and palmitostearins present was considered. We considered it a good risk that, under the conditions of precipitation we used and because of the very small amounts of stearins present, the method of determination was probably much better than any other available. Certainly it is below the limits of the other errors inherent in the experiment.

The values themselves refute Kartha's argument. As he points out, if the tripalmitin were more soluble than the tissue-saturated triglycerides, all the values would be slightly high. Yet the values for endogenous rat fat conform well with Kartha's random theory. and after fat ingestion the values are below those expected by the random theory. It is only the chick fat values that are high, and these are much too high to be accounted for by any possible solubility difference.

b) We were in error in presenting the data in weight percentage instead of mol percentage.

The use of weight instead of mol percentage was made advisedly. To use the mol percentage basis many assumptions and guesses would have to be made as to the fatty acid composition of the various glycerides. The figures would thus be only estimates. The weight percentages could, at least, be given with confidence. More important, the error involved would not be great enough to change any conclusions. The figure of 93% calculated by Kartha in his letter, on assumptions of glyceride fatty acid composition, as compared to our figure of 107%, still leads to the same conclusion that endogenous rat triglycerides are of the random type.

The same is true for the rest of the figures. By whatever method calculated the addition of any fat to a rat diet lowers the percentage of saturated triglycerides below that expected by random distribution.

In the case of chickens the percentage of saturated triglycerides is much greater than expected by random distribution, regardless if calculated on weight or mol percentage basis.

In summary, therefore, while Kartha is technically quite correct in his criticisms of our procedure, these procedures were used advisedly; they are within the limits of error of the experiment; and they are, in our opinion, too small to affect the final conclusions.

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Letter to the Editor

May 12, 1956.

In a recent study of the component acids of salmon egg fat (from *Oncorhynchus gorbuscha*) by R. M. Kyte (1) the author employed fractional distillation of groups of esters segregated by crystallization from acetone. He remarks that the method employed resulted in low values for unsaturation and chain length of the constituent unsaturated fatty acids.

It is true that, unless due precautions are observed, loss of unsaturation in polyethenoid esters is liable