189. *An X-Ray and Thermal Examination of the Glycerides. Part IX. The Polymorphism of Simple Triglycerides.*

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M. p. and *X*-ray data are given for the β′-form of simple triglycerides, which thus exist in four solid modifications and exhibit the same type of polymorphism as the mixed triglycerides discussed in Parts IV, V, VI, and VII (*J.*, 1939, 103, 577, 1141, 1518). The suggestion of Lutton (*J. Amer. Chem. Soc.*, 1945, **67**, 524) and Longenecker and Daubert (*Ann. Rev. Biochem.*, 1945, **14**, 126) that the concept of the glassy (vitreous) state of triglycerides should be eliminated is shown to be untenable.

In Part I (*J.*, 1934, 666) it was shown that simple triglycerides exist in three modifications, namely, the α, β, and vitreous forms; but on extending the study to mixed triglycerides (Parts IV to VII, *locc. cit.*) a fourth form, termed β′, was also observed. This led us to re-examine the simple triglycerides, and we found evidence for the β′-form for those containing odd membered acids (cf. Grüntzig, who reported our results, *Z. anorg. Chem.*, 1939, **240**, 313). At this stage our work was interrupted, but subsequently other workers have reported the expected β′-form for triglycerides containing even-membered acids (Bailey, Jefferson, Kreeger, and Bauer, *Oil and Soap*, 1945, **22**, 10; Lutton, *loc. cit.*; Filer, jun., Sidhu, Daubert, and Longenecker, *J. Amer. Chem. Soc.*, 1946, **68**, 167).

Although these workers obtained *X*-ray evidence for the β′-form, they did not observe any corresponding m. p., neither could they obtain the simple *X*-ray photograph given by the vitreous form. Consequently, Lutton, and Longnecker and Daubert, finding *X*-ray evidence for α, β′, and β forms only, and being aware of only three m. p.s, namely those reported by us for the vitreous, α, and β forms, concluded that we had erroneously associated the *X*-ray patterns and m. p.s. According to them, the three known m. p.s of simple triglycerides are those of the α, β′, and β forms, and the concept of the glassy (vitreous) state of triglycerides should be eliminated.

There are many objections to this view. In the first place, it postulates the existence of only three solid modifications for triglycerides, whereas in Parts IV—VII of this series four modifications have been indubitably established for some forty triglycerides. Again, if our α-m. p.s, which form a non-alternating series, were β′-m. p.s as suggested by Lutton, they would be expected to alternate, since β′-forms of triglycerides are known to possess tilted chains (cf. Malkin, *J.*, 1931, 2796). Moreover, Ravich, Zurinov, Volnova, and Petrov have recently adduced optical and other evidence for the vitreous form of simple triglycerides (*Acta Phytochim.*, 1946, **21**, 101).

Finally, we now report a fourth m. p. (β′) for simple triglycerides, and thus all the saturated triglycerides so far investigated by us exist in four solid modifications, namely, in order of ascending m. p., vitreous, α, β′, and β.

Lutton * states (*loc. cit.*, p. 525): "The conclusive proof of the correspondence of the α-*X*-ray pattern with the lowest melting form, is the experimental result that actual samples which gave the α-*X*-ray pattern also exhibited the lowest form m. p. when tested immediately after *X*-ray exposure".

This evidence, however, is far from conclusive, since both the vitreous and the α-forms give rise to the same side spacing (cf. Figs. 4, 5, Plate), and it is clearly impossible to say whether or not a specimen giving the α-form spacings contains any vitreous form. Indeed, because of the greater crystallinity of the α-form, a specimen preponderantly vitreous would give an *X*-ray photograph very little different from that of an α-form. Again, we have stated in Part I (p. 669) that a number of minute crystalline aggregates are dispersed throughout the vitreous form, and that the exact nature of a solidified glyceride depends very much on the rate of cooling. In our experience, the rods obtained by Lutton by cooling 1 mm. diameter capillaries of molten glyceride from 100° to 0° for "two seconds or longer", would certainly contain varying amounts of α-form. Solid glycerides are extremely bad conductors of heat, and it is difficult to cool the interior of a rod or layer of any appreciable thickness quickly enough to avoid some α-formation. Fig. 5, Plate, was obtained from as thin a layer as possible, cooled rapidly on a cover slip. A specimen treated as stated by Lutton would be expected to melt more or less completely at the vitreous m. p., except for a cloud of α-nuclei which would initiate rapid α-solidification, followed most probably by some α ⟶ β′ transition in the solid state.

Lutton's deductions concerning his beta prime (β′) forms, which gave the softening

* In the following discussion it will suffice to deal with the fuller results of Lutton.

points of our α-forms but the *X*-ray pattern of the β′-forms, are equally debatable, and for the same reason. The strongest lines given by the α- and the β′-forms are practically the same (4·19 and 4·22 respectively), and it is not easy to be certain from an *X*-ray photograph that a β′-form is free from α-form. Our repetition indicates that his β′-forms contained considerable amounts of α-form. Indeed, the fact that Lutton speaks of " softening points " rather than melting points, is itself significant, and confirms this. Thus, all the lower-melting forms are metastable, and transitions into higher-melting forms can take place in the solid state. It is only, therefore, when these transitions are exceptionally slow, *e.g.*, with unsymmetrical mixed triglycerides (Part V), that homogeneous metastable forms can be obtained. Such a form melts to a clear liquid, with all the appearance of a true melting point, and some time may elapse before solidification in the next higher-melting form begins. If, however, owing to transition in the solid state, seeds of the higher-melting form are already present, the lower form melts to a cloudy liquid, which quickly solidifies in the higher-melting form, and, depending on the amount of higher-melting form initially present, the process becomes a softening rather than a melting. In the extreme case, where solid transitions are rapid, and a lower-melting form is full of seeds of higher-melting form, even softening may not be observed, and the m. p. may escape observation. This is the case with simple triglycerides of even acids, and considerable patience and repetition is required to determine the β′-m. p., which is best done as follows. Melt the specimen in a capillary tube, solidify it in ice, and then place it in a bath one or two degrees above the m. p. of the vitreous form. Complete or nearly complete melting takes place, followed closely by resolidification in the α-form, but after some three to six minutes longer at this temperature, a further change takes place, denoted by a shrinking from the walls of the capillary. Before this change the specimen melts almost completely at the α-m. p., but, after the shrinking, the α-m. p. is passed without any change in appearance until the β′-m. p. is reached. At this point, softening and very rapid resolidification in the stable β-form takes place.

In contrast to the above, transitions of glycerides of odd acids are much slower, and little difficulty is experienced in determining their β′-m. p.s.

Table I gives our latest m. p. data for simple triglycerides.

TABLE I.

	β.	β′.	α.	Vitreous.
Tristearin	72·0°	70·0°	65·0°	54·5°
Trimargarin	64·0	62·5	61·0	50·0
Tripalmitin	65·5	63·5	56·0	45·0
Tripentadecylin	54·0	53·0	51·5	40·0
Trimyristin	57·0	54·5	46·5	33·0
Tritridecylin	44·0	42·5	41·0	25·0
Trilaurin	46·4	—	35·0	15·0
Triundecylin	30·5	29·0	26·5	1·0
Tridecylin	31·5	—	18·0	−15·0

β′-M. p.s could not be observed for trilaurin and tridecylin.

X-*Ray Investigation.*—β′-Forms were obtained as described above, and side and long spacings determined. Owing to the rapid transition β′ ⟶ β for triglycerides of even acids, the lines of both these forms are usually present on the same photograph (cf. Fig. 8, Plate) where the inner strong line is the strongest line of the β-spacing (4·6 A.), the remaining two strong lines being the β′-spacings of 4·22 and 3·8 A. (Lutton, *loc. cit.*, gives 4·18 and 3·78 A.). No difficulty is experienced, however, in obtaining homogeneous β′-forms of odd acid triglycerides, as these can be kept for some time just below the β′-m. p. without transition into the stable form. By what appears to be more than a coincidence, the significance of which still eludes us, the side spacings of tritridecylin, tripentadecylin, and trimargarin are identical with those of the β-forms of β-myristodilaurin, β-palmitodimyristin, and β-stearodipalmitin respectively, the total chain lengths in the respective pairs being the same (cf. Figs. 13, 12, 11, Plate, with Figs. 11, 12, 13 on Plate facing page p. 107, *J.*, 1939 *). Long spacings of β′-forms of odd acid triglycerides are, within 0·2 A., the same as those of the β-forms.

As indicated above, the American workers have been misled in interpreting their *X*-ray data, because of the difficulty of obtaining homogeneous metastable forms, and whilst, at first glance, their results appear to support their view, it is not difficult to show that this is untenable. If, *e.g.*, the lowest-melting form of tristearin (m. p. 54·5°) is in fact the α-form, it

* The value, 4·48 A., given there for β-stearodipalmitin should be corrected to 4·38.

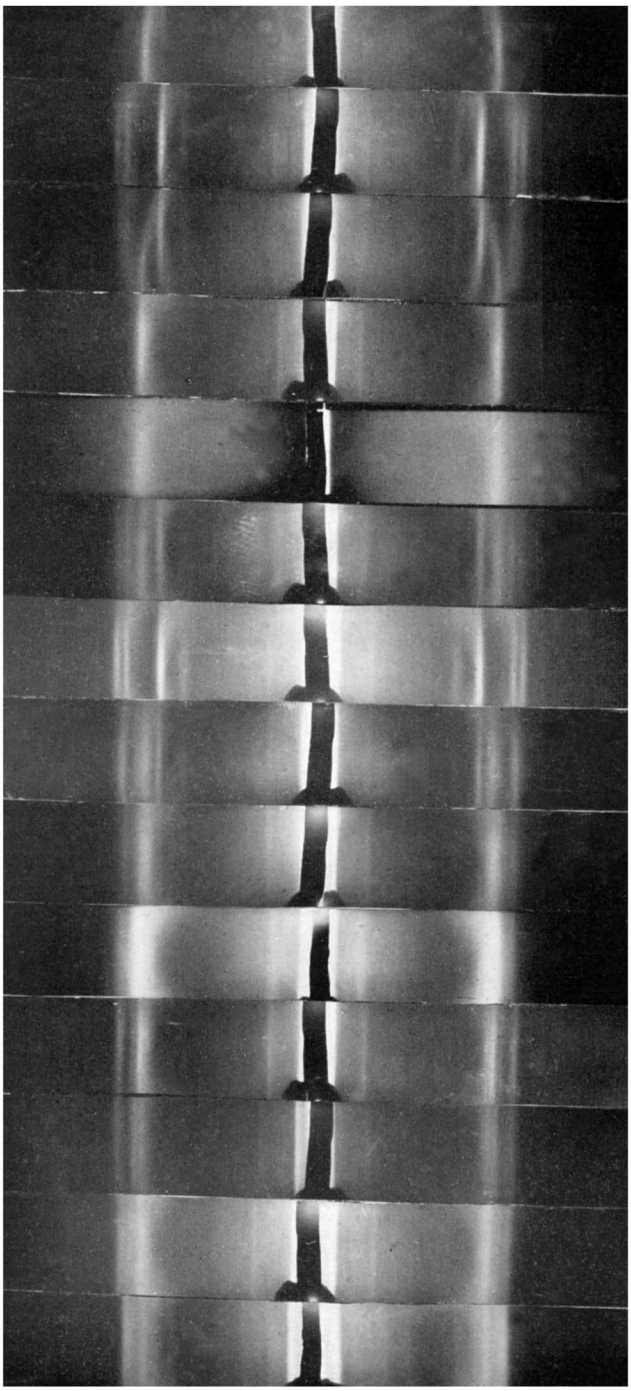

FIG. 1 — *Tristearin, mainly α, beginning to change to β.*

2 — *Tristearin, β.*

3 — ,, *α and β.*

4 — ,, *α.*

5 — ,, *vitreous.*

6 — ,, *α and β.*

7 — *Tripalmitin, β.*

8 — ,, *β′ and β.*

9 — ,, *α and β′.*

10 — *Trimargarin, α and β.*

11 — ,, *β′.*

12 — *Tripentadecylin, β′.*

13 — *Tritridecylin, β′.*

14 — *Triundecylin, β′.*

[*To face p.* 986

TABLE II.

Long and side spacings of β′-forms of simple triglycerides.

	Long.	Side.			
Tristearin	47·2	3·8	4·22		
Tripalmitin	42·6	,,	,,		
Trimyristin	37·6	,,	,,		
Trimargarin	43·7	3·79s	4·02w	4·19s	4·37w
Tripentadecylin	39·1	3·81s	4·12s	4·3s	
Tritridecylin	34·2	3·85m	4·04w	4·26s	4·43w
Triundecylin	29·5	3·88s	4·25s	4·52w	

	Lutton.			Bailey, *et alia*.		
Tristearin	46·8	3·78	4·18	46·4	3·79	4·19
Tripalmitin	42·3	,,	,,			
Trimyristin	37·65	,,	,,			

s = strong; m = moderate; w = weak.

should be impossible to obtain an *X*-ray photograph, showing the single side spacing of this form, at any temperature above 54·5°. Figs. 1 and 3 are photographs of a rod of tristearin, initially in the α-form, surrounded by an oven at a rising temperature from 57° to 64°. Fig. 1 shows the α-form at the top of the rod beginning to change to the stable β-form, whilst Fig. 3 (64°) shows the specimen mainly in the β-form, but still in the α-form in the lower part of the rod which is at a slightly lower temperature owing to contact with the metal spectrograph. (The curvature of the lines in this photograph is due to the fact that the specimen is now a shortened rod of β-form on top of a very short rod of α-form, instead of a uniform rod of normal length.) Again, Fig. 6 is a photograph of a rod of tristearin, originally α-form, after being in a thermostat at 60° for ½ hour. According to the American workers the α-form could not exist at this temperature, yet both α- and β-forms are present.

Repetition of Lutton's work leaves no doubt that his β′-forms contained varying amounts of α-form. A rod of tripalmitin made by his method for obtaining β′-forms, *i.e.*, melting the the specimen at 100°, cooling it in ice, and then keeping it at 45° for a minute or two, gave the photograph, Fig. 9, showing a strong diffuse inner line and a weaker sharper line. This is clearly a mixture of α- and β′-forms, the former predominating, since the true β′-form gives two lines of equal intensity (cf. Fig. 8). Moreover, by keeping the specimen for a longer period at 45°, or by increasing this temperature slightly, it is possible to increase the relative intensity of the weaker line, and the strong diffuse line becomes better defined as the α-line disappears. This is seen much more clearly with odd acid glycerides, where the β′-form gives a more definite and characteristic photograph uncomplicated by the β′ ⟶ β transition. By treating trimargarin according to Lutton's method for obtaining β′-forms one obtains Fig. 10, showing again the strong α-line overlaid by the weaker β′-lines, very similar, indeed, to the palmitin photograph. On raising the temperature of the specimen to just below the β′-m. p., the spacings of the β′-form alone are obtained (Fig. 11).

Our present results complete the broad picture of the polymorphism of saturated triglycerides. All that we have examined, whether simple or mixed, exist in four solid forms, *viz.*, vitreous, α, and β′ (crystalline, monotropic), and β (stable crystalline form). Except for those containing short chains (*i.e.*, C_{10}, C_{12}) rapid cooling (ice) brings about solidification mainly in the vitreous form, and this, on warming, undergoes the transitions α ⟶ β′, β′ ⟶ β, and also in some cases α ⟶ β. These transitions are rapid for simple even acid, and symmetrical mixed triglycerides, and slow for simple odd acid, and unsymmetrical mixed triglycerides, the speed of transitions in all cases diminishing with increasing length of acid chain. Also, as would be expected, impurities decrease the rate of transitions. It is of interest to note that α-forms of simple even acid triglycerides may change directly into the stable β-form without passing through the intermediate β′-phase, and this may, of course, be true for other glycerides, although we have not observed it. The factors determining which transition takes place are not easy to determine, but in general α ⟶ β transition appears to be more probable the purer the glyceride and the smaller the amount of vitreous form present with the α-form on solidification.

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