Nucleation Behaviour of Tripalmitin from a Triolein Solution

W.L. Ng
Department of Chemistry, University of Malaya, 59100 Kuala Lumpur, Malaysia

Nucleation of tripalmitin (PPP) crystals from melt and from its binary triolein (OOO) solutions was studied by differential scanning calorimetry (DSC) and optical microscopy. Cooling DSC thermograms for either pure PPP or OOO exhibits a single exotherm. For the solutions, two widely separated peaks due to PPP and OOO were observed. The PPP peak is broadened and shifts down to a lower temperature as the OOO content in the solution increases. A modified Turnbull and Fisher equation was used for nucleation data analysis. The temperature for maximum supercooling of the solution is reduced by OOO but the degree of maximum supercooling remains unchanged at 22°K. An interfacial free energy of 9.45 erg/cm^2 for β-PPP/ melt was deduced. This value drops rapidly to a slightly lower value of 8.95 erg/cm^2 in triolein solution. The effect of a reduced interfacial free energy on nucleation is critically discussed in the light of a simultaneous reduction in the melting point of β-PPP in the solution.

Many of the important properties of vegetable oils and animal fats are controlled by phase transitions (1) of their component triglycerides. These include solid-liquid (melting), liquid-solid (crystallization) and solid-solid (aging or polymorphic) transitions. Previous studies were focused on the description of polymorphism of individual triglycerides. (2–4). It is now generally accepted that simple triglyceride crystallizes in three (5) polymorphic forms: α, β′ and β in increasing order of melting point and thermal stability. There are two β′ polymorphs (designated as β'_1 and β'_2) for certain triglycerides (6). The investigation on triglyceride interactions (5,7) and polymorphism (8) in binary systems is relatively recent. Of late, there has been a growing interest on the kinetics of crystallization of triglycerides from solution (9), melt (10,11) and with surfactant additives (11). However, no crystallization work has been reported on binary or multicomponent systems which closely resemble the composition of oils and fats. Phase behaviour in such systems is often complicated by mutual interactions among the component triglycerides. Besides exhibiting intersolubility, the effect of triglyceride interactions (12) may result in the formation of (a) solid solutions (mixed crystal), (b) eutective mixtures and (c) chemical compounds.

The binary system of greatest interest for many industrial processes of oils and fats is one in which the melting points of the component triglycerides lie on opposite sides of ambient temperature (8). At room temperature, tripalmitin (PPP) is a simple saturated solid whereas triolein (OOO) is a simple, unsaturated, liquid triglyceride. Only the melting behaviour of this binary system has hitherto been investigated. It was found that the melting point (8,13) of pure β-PPP crystals in contact with their OOO solution was lowered and the enthalpy of melting (8) was affected by the composition of the melt. The fact that OOO modifies the melting behaviour of PPP indicates unequivocally the existence of intermolecular interaction at the crystal/solution interface. Such interaction might also modify its interfacial free energy and henceforth affect the rate of formation of crystal nucleus. In this work, differential scanning calorimetry (DSC) and optical microscopy were used to study crystallization behaviour and nucleation of PPP in a OOO solution and to determine the associated nucleation parameters.

EXPERIMENTAL PROCEDURES

E.P. grade (> 95%) tripalmitin and triolein from Tokyo Kasei Kogyo Co. were used without further purification. Binary solutions of PPP and OOO were prepared at 343°K with stirring for 30 min to ensure complete mixing. A tiny drop of the sample was then transferred while hot by microliter syringe to the centre of a clean, circular, microscope glass cover slip (19mm dia. × 0.21 mm) which was then covered by another slip forming a thin film of oil sample (approx. 2mm dia.) in between. The edges of the slips were sealed with fast setting epoxy resin. The sandwiched sample was heated inside an oven at 343°K for 30 min to destroy any existing nuclei before it was transferred quickly onto a hot stage attached to a polarizing microscope. The hot stage was maintained at the desired temperature by circulating a water/glycol mixture from a thermostated bath (±0.1°K). The temperature of the sample was monitored by a built-in thermometer inside the hot stage. The polarizer was rotated such that the liquid sample appeared dark when viewed through the eyepiece. When nucleation occurred, the crystals appeared bright against the dark background so that it was easily detected even though it was rather small when first formed. The overall magnification used was 100 ×. The induction time τ is the time taken for the appearance of the first nucleus after the sample disk has been properly placed onto the hot stage. A Perkin-Elmer DSC-2 differential scanning calorimeter complete with a data station, and a TADS-1 plotter was used to obtain a crystallization thermogram of the sample at a cooling rate of 5°K/min from 343°K to 233°K except for OOO which was cooled from 290° to 213°K. This was followed by the heating of the sample at a rate of 5°K/min to the starting temperature to obtain its melting thermogram. About 5–10 mg of the sample was used for each run. The melting point was determined using a 75 mm × 1 mm diameter capillary tube. The tube which contained about 1 cm of sample was frozen at 227°K for 16 hours. It was then suspended in a water bath which was heated at a rate of 1°K/min initially and reduced to 0.5°K/min on approaching the expected melting point. The temperature at which the solid completely melted was taken to be its melting point.

RESULTS AND DISCUSSION

Crystallization Behaviour. Crystallization consists of three consecutive stages: (a) supersaturation or supercooling, (b) nucleation and (c) crystal growth. A melt or solution will rarely undergo nucleation unless it has been supercooled sufficiently. All samples (with the exception of OOO) were initially tempered at 343°K for 30 min prior to cooling in order to destroy any nuclei that might persist in the solution or melt. For either pure PPP or OOO their solidification thermograms (curves *a* and *b* in Fig. 1) show only a single exotherm at 317°K and 233°K respectively. This means that only one polymorph was formed when these individual simple triglycerides were cooled at 5°K/min to 230°K. On the other hand, in the binary solutions two widely separated peaks were observed. The high temperature peak is slightly below that of pure PPP whereas the low temperature peak is higher than that of pure OOO. It is interesting to note that these separate exotherms are composition dependent and both peaks shift to lower temperature as the OOO content in the solution increases.

As the temperature was lowered at a prescribed rate, the solution was brought from a stable state of unsaturation into a labile state of supersaturation with an accompanying increase in the degree of supercooling. Once nucleation occurs, excess solute (PPP) from the supersaturated solution begins to crystallize out until its concentration falls back to its equilibrium value (saturation). This continuous precipitation of solid PPP as the solution is being cooled causes a broadening in the crystallization exotherm of PPP as seen in curve Fig. 1*c*. Such solubility effect on the thermogram was absent in the pure tripalmitin melt (curve Fig. 1*a*).

With increasing OOO content in the solution, the thermogram depicts a doublet PPP exotherm (curves Fig 1*d* and *e*) with the second peak only a few degrees below the first. Because there is no eutectic mixture or mixed crystal formation in this system (13), this doublet must be attributed to two different polymorphs of PPP. One of them previously has been identified to be a β crystal (8). The order of occurrence suggests that the lower-temperature exotherm is either the α- or β'-polymorph of PPP which has a lower melting point.

The polymorphic nature of the crystals can be revealed by reference to their melting thermogram. Figure 2 displays DSC thermograms of three systems upon heating them at 5°K/min: (a) PPP, (b) OOO and (c) 51% PPP. In system *a*, the larger peak at 337°K is due to β-PPP and the smaller one at 317°K is α-PPP. The initially formed α-crystal must have undergone partial $\alpha \rightarrow \beta$ polymorphic transformation during the heating process. In system *b* only a single peak at 266°K occurs and this corresponds to β'_2-form of OOO (T_m = 265°K) (14). In the binary system *c*, the solution completely solidifies at 230°K into a mixture of PPP (β + other polymorphs) and OOO (β'_2-form). During subsequent heating, β'_2-OOO melts at 266°K and PPP remains in the solid phase (curve *c*). As the triglycerides are not mutually soluble in the solid state, the melting point of β'_2-OOO is thus not affected at all by the presence of PPP (Table 1) in the solid. At higher temperatures there is a single endotherm due to the β polymorph of PPP as has been confirmed by x-ray diffraction measurements (8). As there is no sign of a second melting endotherm other than the one for β-PPP, the initially formed α- or β'-PPP could have easily dissolved in OOO due to their lower melting points or have undergone polymorphic transformation to the β-form during the process of heating them.

When the solution was sufficiently supercooled to bring about nucleation, the heat evolved causes an initial rise of the exotherm. The position of the peak is therefore closely related to the crystallization temperature of the solution. The up-shift of the β'_2-OOO peak with increasing PPP content implies that nucleation of OOO in the binary system is heterogeneous in nature. Nucleation is induced by the previously formed PPP crystals which causes a corresponding reduction

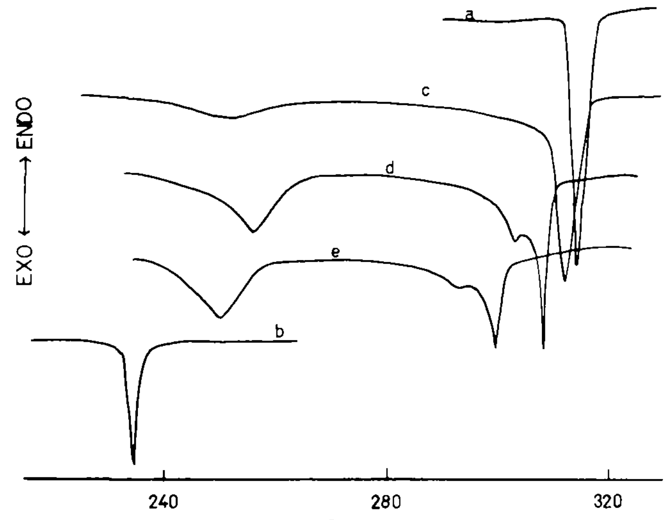

FIG. 1. Crystallization DSC thermograms of a) PPP, b) OOO, c) 71.0%, d) 51.0%, e) 20.5% PPP in triolein solutions. Vertical axis not to scale.

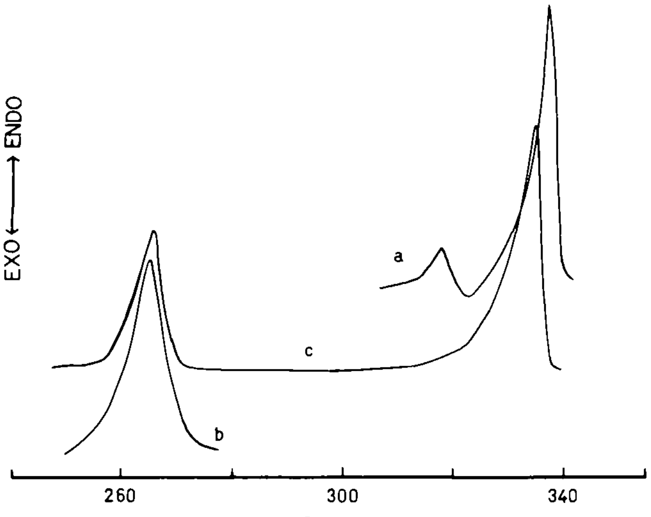

FIG. 2. Melting DSC thermograms of a) PPP, b) OOO, c) 51.0% PPP in triolein solution. Vertical axis not to scale.

TABLE 1

Solid ↔ Liquid Transition Temperatures from DSC Thermograms

PPP	Crystallization, T,°K		Melting, T,°K	
mole%	Triolein	Tripalmitin	Triolein	Tripalmitin
100.0		317		318, 337
71.0	253	310	266	335
51.0	256	302, 308	266	334
20.5	250	292, 300	266	330
0	233		266	

in the degree of supercooling. Thus, crystallization occurs at a higher temperature closer to its melting point of 266°K. On the other hand, the lowering of the crystallization temperature (down-shift of the peak) of β-PPP is likely to be the result of an intermolecular interaction between PPP and OOO molecules at the crystal/solution interface. This interaction causes a similar reduction in the melting point of β-PPP.

Crystal/Melt Interfacial Free Energy. A melt below its melting point, T_m, can decrease its free energy by undergoing nucleation. This is because the solid phase has a lower free energy than the liquid. However, formation of solid nucleus is always accompanied by the simultaneous creation of solid-liquid interface which increases the free energy by an amount (the interfacial free energy) for each unit area of the interface. Thus, a supercooled liquid can even remain remarkably stable without undergoing crystallization unless nucleation can result in a decrease of the overall free energy of the system. For a spherical nucleus of radius *r*, the surface free energy contribution is $4\pi r^2\sigma$ and the bulk free energy contribution is $\frac{4}{3}\pi r^3\,\Delta G_v$ where $\Delta G_v = \Delta H(T_m - T)/vT_m$ is the bulk free energy change per unit volume *v* of the solid nuclei, ΔH is the enthalpy of melting (15). The overall free energy change upon nucleation is therefore given by:

$$\Delta G = -\tfrac{4}{3}\pi r^2\,\Delta G_v + 4\pi r^2\sigma \qquad [1]$$

There exists a critical radius $r_c = 2\sigma/\Delta G_v$ above which, if the nucleus grows, the total free energy is decreased. The corresponding critical free energy is the activation free energy of nucleation:

$$\Delta G^c = \frac{16\pi r^3}{3(\Delta G_v)^2} = \frac{16\pi\sigma^3 v^2 T_m}{3\Delta H^2\,(Tm-T)^2} \qquad [2]$$

Thus, there is a thermodynamic potential barrier in creating a critical nucleus. Once the local fluctuation in energy is sufficiently large to overcome this barrier, spontaneous nucleation will then occur. According to Buckle's analysis (16) of the Turnbull and Fisher equation, the induction time (τ) of nucleation is:

$$\tau = \frac{1}{\sqrt{4\sigma kT}}\,v^{-1/3}h\,\exp\,(\Delta G_d + \Delta G^c)/kT \qquad [3]$$

In the formula these symbols are used: ΔG_d (the activation free energy for the incorporation of a molecule into the crystal nucleus) and v (molecular volume of the solid). The other symbols have their usual meanings.

For equation [3] to be applicable, nucleation must be carried out isothermally. The solution was first supercooled to the desired temperature below the melting point of β-PPP (the highest melting polymorph) and held constant at the temperature for nucleation to occur. It has been found previously that the β-polymorph of PPP is exclusively crystallized out from the triolein solution (8) and from pure PPP melt above 323°K (11). The temperature for maximum cooling, T_s, is the lowest temperature to which a system can be supercooled with a corresponding induction time (τ) arbitrarily fixed at 10s (17). This can be determined graphically by extrapolating from a plot of log τ versus T to intercept at the line log τ=1 (Fig. 3). Equation [4] as follows, is derived by rearranging equation [3], putting $T = T_s$, $\tau = 10s$ and $\Delta G_d = 30.4$ kJ/mol (18) and substituting numerical values for the physical constants:

$$19.55 + \tfrac{1}{3}\log v + \tfrac{1}{2}\log T_s - 1589/T_s = 5.279\times10^{16}\,\frac{\sigma^3 v^2\,T_m^{\,2}}{\Delta H^2(T_m - T_s)^2 T_s} \qquad [4]$$

Equation [4] was then used to determine σ by means of numerical analysis using the corresponding values given in Table 2. The calculated interfacial free energy σ for β-PPP/melt is 9.45 erg/cm². This value compares favourably with the value of 8.3 erg/cm² reported for the α-PPP/melt (17). In OOO solution, the interfacial free energy for β-PPP drops rapidly to 8.95 erg/cm² (Table 2). This crystal/solution interfacial free energy σ is made up of (19):

$$\sigma = \sigma_c + \sigma_s - U \qquad [5]$$

where σ_c and σ_s are respectively surface energies of the crystal and of the solution and U is the interaction energy. The OOO molecules which are adsorbed at the crystal/solution interface must have interacted with the PPP molecules to bring about a reduction in the interfacial free energy. This intermolecular interaction may be composed of polar or dispersive forces. An

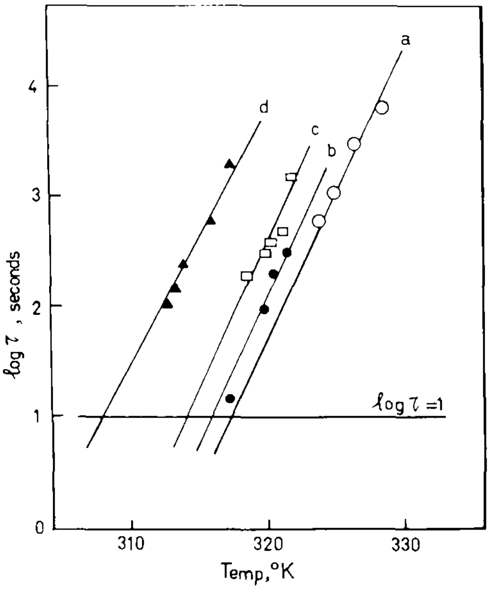

FIG. 3. Plot of log τ, induction time versus T, temperature of crystallization for a) PPP, b) 71.0%, c) 51.0%, d) 20.5% PPP in triolein solutions. Interceptions on the line log τ = 1 give the respective temperatures for maximum supercooling T_s.

TABLE 2

Nucleation Parameters of PPP from its 000 Solutions

PPP mole%	T_s,°	T_m,°K	$10^{12}\Delta H^*$ erg/ molecule	10^{21} v† cm/ molecule	σ erg/cm^2	ΔG^c kJ/mol	ΔG^c(317K) kJ/mol
100.0	317.1	339.2	2.823	1.502	9.45	57.0	57.0
71.0	315.6	337.7	2.648	1.500	9.06	56.1	64.6
51.0	314.0	336.2	2.585	1.499	8.95	55.7	75.3
20.5	307.0	329.2	2.567	1.491	8.95	53.7	180.7

*: calculated from the data in ref. 8.
†: calculated using the density data in ref. 18.

immediate effect arising from a smaller interfacial free energy of β-PPP/solution is a reduction of the activation free energy of nucleation. However, nucleation is retarded by the smaller degree of supercooling (T_m −T) caused by the concurrent lowering of the melting point of β-PPP in the solution. This retarding effect is so great that it outweighs the catalytic effect of the reduced interfacial free energy and hence causes a drastic increase in the activation free energy of nucleation at any temperature, e.g. 317°K as calculated from equation [2]. This implies that nucleation of β-PPP is rendered more sluggish and difficult to occur in a OOO solution than in its own melt. On the other hand, if nucleation is carried out at the respective T_s of the solutions such that the maximum degree of supercooling (T_m − T_s) remains constant at 22°K, then the catalytic effect of a reduced interfacial free energy becomes noticeable. The calculated activation free energy at the respective temperature of maximum supercooling $\Delta G^c(T_s)$ indeed shows a steady decrease (Table 2) with an increasing OOO content in the solution studied.

REFERENCES

1. Knoester M., P. de Brieijne and M. van den Tempel, *Chem. Phys. Lipids 9*:309 (1972).
2. Duffy, P.J., *J. Chem. Soc. 5*:197 (1853).
3. Malkin T., and J. Clarkson, *J. Chem. Soc. 666*:985 (1934).
4. Lutton E.S., *J. Amer. Chem. Soc. 67* (1945) 524.
5. Small, D.M., in *Handbook of Lipid Research*, edited by D.J. Hanahan, vol. 4, Plenum, New York, 1986, p. 345.
6. Simpson T.D., *J. Am. Oil Chem. Soc. 60*:95 (1983).
7. Rossell J.B., *Advances in Lipid Research 5*:353 (1967).
8. Norton I.T., C.D. Lee-Tuffnell, S. Ablett and S.M. Bociek, *J. Am. Chem. Soc. 62*:1237 (1985).
9. Albon, N., D. Illingworth and R. Hull, *J. Cryst. Growth 2*:26 (1968).
10. Skoda, W., and M. Tempel, *J. Cryst. Growth 1*:207 (1967).
11. Sato, K., and Kuroda T., *J. Am. Oil Chem. Soc. 64*:124 (1987).
12. Rossell J.B., *Chem. Industry 1*:832 (1973).
13. Hale J.E., and F. Schroeder, *Lipids 16*:805 (1981).
14. Hagemann J.W., and W.H. Tallent, *J. Am. Oil Chem. Soc. 49*:118 (1972).
15. Brice, J.C., in *The Growth of Crystals from Liquids*, North Holland, Amsterdam, London, 1973, p. 78.
16. Buckle, E.R., *Proc. Roy. Soc. A, 261*:189 (1960).
17. Phipps L.W., *Trans. Faraday Soc. 60*:1873 (1964).
18. Phipps L.W., *Nature 193*:541 (1962).
19. Walton A.G., in *Nucleation*, edited by A.C. Zettlemoyer, Marcel Dekker, New York, 1969, p. 225.

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