Anionic Surfactant Effect on Viscosity of Salicylic Acid–Cetrimide System

LUCY S. C. WAN

Abstract □ Some anionic surfactants, sodium lauryl sulfate, sodium alkyl aryl polyether sulfate, dioctyl sodium sulfosuccinate, and sodium alkyl aryl polyether sulfonate, were found to increase the viscosity of the salicylic acid–cetrimide system to a maximum and then decrease it on further addition of the surfactant solution. The viscosity rise could be due to the attraction of free cetrimide molecules for the anionic surfactant, which would result in the removal of the cationic surfactant from the dispersion medium. This probably led to the approach of the macromolecules formed by the interaction of salicylic acid with cetrimide closer together and, thus, increased the viscosity. When an adequate amount of the anionic surfactant is added, most of the free cetrimide molecules would be neutralized and a slight excess of negatively charged surfactant ions could be present. Under such conditions, there could be a disruption of the mesh structure, rendering the system unstable and resulting in a viscosity reduction.

Keyphrases □ Surfactants, anionic—effect on salicylic acid–cetrimide system viscosity □ Salicylic acid–cetrimide system—effect of anionic surfactants on viscosity □ Cetrimide–salicylic acid system—effect of anionic surfactants on viscosity □ Viscosity—role of anionic surfactants in salicylic acid–cetrimide system

From previous findings (1), it appeared that the macromolecules produced by the interaction of salicylic acid and cetrimide were sensitive to several types of additives. Since surfactants were involved in this interaction, the addition of surface-active materials seemed to be appropriate in the determination of its stability toward substances with which the compounds shared a common property, that of being surface active. Isemura *et al.* (2) showed that some anionic and cationic surfactants interacted with gelatin and that the charge on the surfactant and the length of the chain affected the binding to the gelatin. Apparently, no other formal study of this kind has been made.

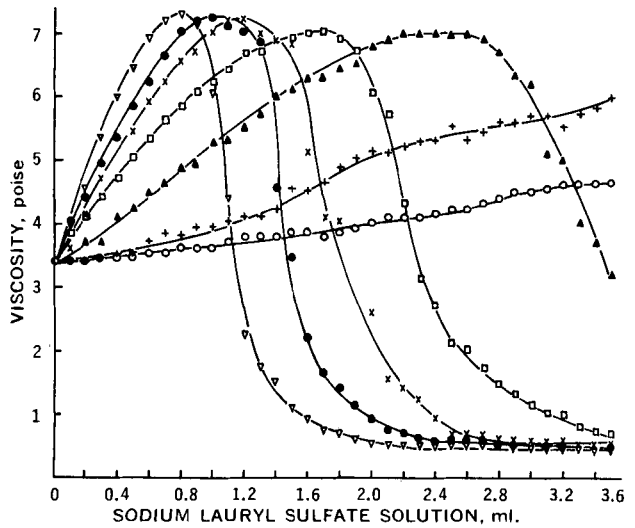

Figure 1—*Effect of varying concentrations of sodium lauryl sulfate on the viscosity of systems containing 5% cetrimide and 1.4% salicylic acid at 25°. The sample weight is 50 g. and the shear rate is 78.56 sec.$^{-1}$. Key: sodium lauryl sulfate concentration (%): ▽, 12; ●, 10; ×, 8; □, 6; ▲, 4; +, 2; and ○, 1.*

Table I—Comparison of Amounts of Various Anionic Surfactants which Cause a Viscosity Decrease, after the Maximal Viscosity Has Been Attained, of Systems Containing 1.4% Salicylic Acid and 5% Cetrimide at 25°

Concentration of Surfactant Solution Added, %	Weight of Surfactant (g.) to Reduce Viscosity of Salicylic Acid–Cetrimide System (50 g.) Initially			
	Sodium Lauryl Sulfate	Dioctyl Sodium Sulfosuccinate	Sodium Alkyl Aryl Polyether Sulfate	Sodium Alkyl Aryl Polyether Sulfonate
2	0.096	0.088	—	—
4	0.108	0.084	0.148	0.132
6	0.108	0.084	0.132	0.144
8	0.104	0.088	0.144	0.136
10	0.110	0.100	0.160	0.150
12	0.108	—	—	0.144

EXPERIMENTAL

Materials—The anionic surfactants used were sodium lauryl sulfate[1], sodium alkyl aryl polyether sulfate[2], dioctyl sodium sulfosuccinate[2], and sodium alkyl aryl polyether sulfonate[2]. With the

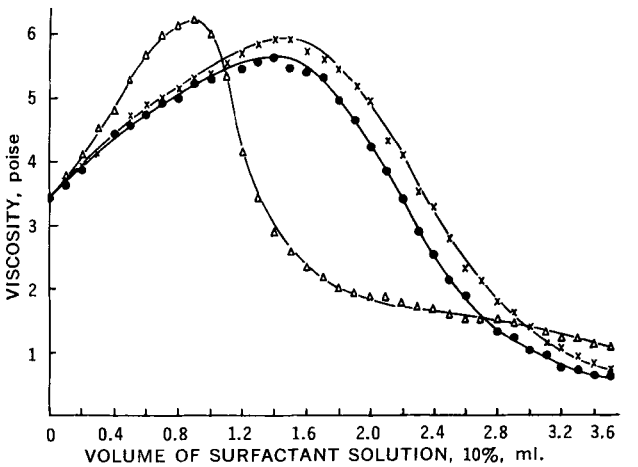

Figure 2—*Effect of solutions of anionic surfactants (10%) on the viscosity of systems containing 5% cetrimide and 1.4% salicylic acid at 25°. The sample weight is 50 g. and the shear rate is 78.56 sec.$^{-1}$. Key: ●, sodium alkyl aryl polyether sulfonate; ×, sodium alkyl aryl polyether sulfate; and △, dioctyl sodium sulfosuccinate.*

[1] Sipon Products Ltd., London, England.

[2] Marketed as Triton X-301, Triton GR-5, and Triton X-200, respectively, by Rohm & Haas Co., Philadelphia, PA 19105, to whom the author is very grateful for the samples supplied. The active ingredient content in Triton X-301, Triton GR-5, and Triton X-200 is 20, 60, and 28% w/w, respectively.

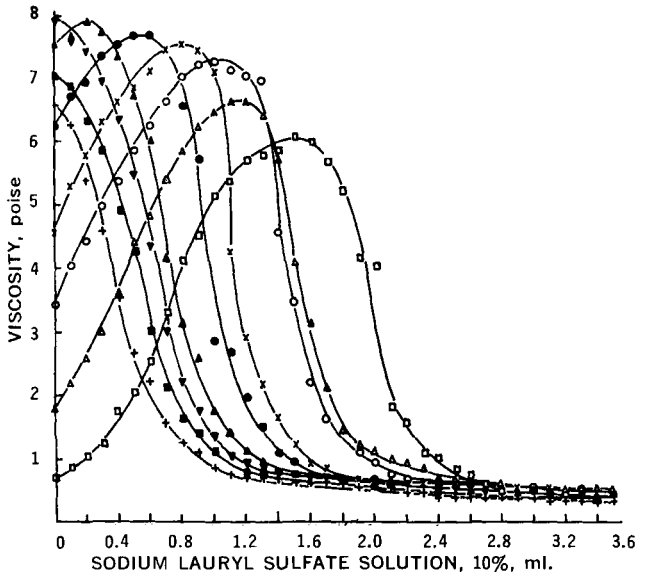

Figure 3—*Viscosity change in systems containing 5% cetrimide and different salicylic acid concentrations in the presence of sodium lauryl sulfate solution (10%) at 25°. The sample weight is 50 g. and the shear rate is 78.56 sec.$^{-1}$. Key: salicylic acid concentration (%): □, 1.2; △, 1.3; ○, 1.4; ×, 1.5; ●, 1.6; ▲, 1.8; ▼, 1.9; ■, 2.0; and +, 2.1.*

exception of sodium lauryl sulfate, the surfactants were in the form of dispersions, with the strength of the active ingredient expressed on a weight percentage basis. Hence, all surfactant solutions used as additives were prepared on this basis. Cetrimide BP[3] and recrystallized salicylic acid, m.p. 158–159°, were the same as those mentioned in a previous paper (1).

Measurement of Viscosity at 25°—The method described earlier (1) was adopted[4].

RESULTS AND DISCUSSION

Figure 1 shows the effect of the addition of sodium lauryl sulfate solutions of different concentrations on the viscosity of a system containing 5% cetrimide and 1.4% salicylic acid. This system is the same as that used in a previous study (1). The viscosity increases with the volume of additive up to a maximum value and decreases thereafter. The rise and fall in viscosity occur with different sodium lauryl sulfate concentrations except with low surfactant concentrations. Only after the addition of 4.7 ml. of sodium lauryl sulfate solution is there a reduction in viscosity. With the 1% surfactant solution, even after incorporating 8 ml., there is no indication of a fall in viscosity. Similar changes in viscosity are also produced by sodium alkyl aryl polyether sulfate, dioctyl sodium sulfosuccinate, and sodium alkyl aryl polyether sulfonate (Fig. 2). The concentration range used was the same as that employed with sodium lauryl sulfate. Although the initial decrease in viscosity takes place at different volumes of the additive, the calculated weight of sodium lauryl sulfate required to bring about this reduction is almost constant (Table I), irrespective of the concentration of sodium lauryl sulfate used. Similar results are obtained with the other anionic surfactants (Table I).

The rise and fall in viscosity are largely due to the effect of the attraction of the anionic surfactant for the cationic ion in the system. It was shown previously (3) that salicylic acid interacted with cetrimide to produce a marked change in viscosity, which is believed to be brought about by the formation of macromolecules. It is thought that the macromolecules are linked together in a network. This arrangement can increase the viscosity of the solution and the size of the macromolecules. In the system containing 5% cetrimide and 1.4% salicylic acid, not all of the cetrimide molecules interact with the acid. When more salicylic acid is added, in the absence of the additive (Fig. 3), the viscosity increases still further, up to a point just below or very near saturation, *i.e.*, a system containing 1.9% acid. When it is above saturation, namely 2.0% and greater, the system becomes less viscous. This is taken as an indication of instability. This also shows the importance of the presence of free cetrimide molecules in the dispersion medium, *i.e.*, molecules of surfactant that have not interacted with the acid. Their presence provides the positive charge to the dispersion and confers stability.

On the initial addition of sodium lauryl sulfate solution, the free cetrimide molecules are attracted to the added anionic surfactant but some free cetrimide molecules remain in the dispersion medium. If sufficient amounts are added, the anionic surfactant forms a precipitate of the hydrophobic anion with the hydrophobic cation. The presence of the precipitate tends to increase the viscosity by increasing the content of the dispersed particles or, possibly, by expanding the network of macromolecules by interpenetration. These effects, however, are considered to be minor on the viscosity change. As can be seen from Fig. 1, the greater the concentration of sodium lauryl sulfate, the higher is the viscosity rise. Cloudiness and eventually precipitation appear at different stages during the addition of the anionic surfactant. A precipitate still remains after the viscosity decreases. With low concentration of the additive, the viscosity increases only to a small extent due to the inadequate

Table II—Relationship between Amount of Surfactant Required to Decrease Viscosity Initially and Salicylic Acid Concentration of Systems Containing 5% Cetrimide and Different Acid Quantities[a]

Salicylic Acid Concentration, %	Weight of Surfactant (g.) which Decreases Viscosity Initially: Sodium Lauryl Sulfate	Sodium Alkyl Aryl Polyether Sulfate	Sodium Alkyl Aryl Polyether Sulfonate	Dioctyl Sodium Sulfosuccinate
1.2	0.160	0.270	0.220	0.150
1.4	0.110	0.160	0.150	0.100
1.6	0.070	0.080	0.090	0.060
1.8	0.030	0.020	0.020	0.010
1.9	0.010	0.010	0.010	0.010

[a] Sample weight = 50 g., temperature = 25°, and shear rate = 78.56 sec.$^{-1}$.

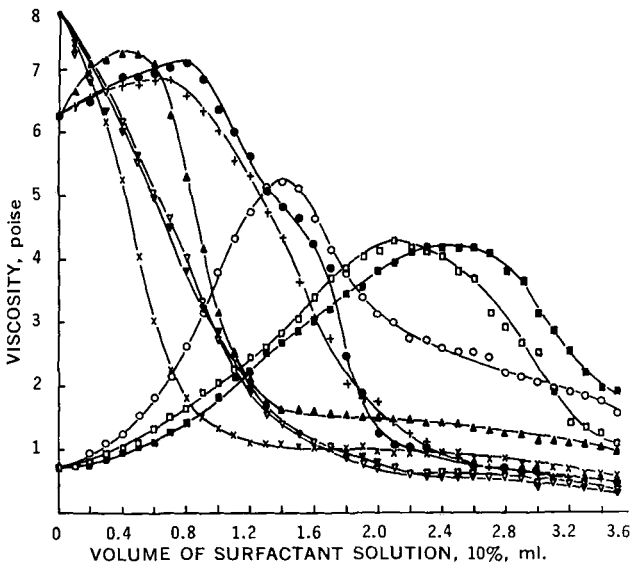

Figure 4—*Viscosity change in systems containing 5% cetrimide and different salicylic acid concentrations in the presence of solutions of anionic surfactants (10%) at 25°. The sample weight is 50 g. and the shear rate is 78.56 sec.$^{-1}$. Key: salicylic acid concentration (%): for sodium alkyl aryl polyether sulfonate—▽, 1.9; ●, 1.6; and □, 1.2; for sodium alkyl aryl polyether sulfate—▼, 1.9; +, 1.6; and ■, 1.2; for dioctyl sodium sulfosuccinate—×, 1.9; ▲, 1.6; and ○, 1.2.*

[3] Glovers Chemicals Ltd., Leeds 12, England.
[4] A portable Ferranti viscometer, model VL, was used in this study.

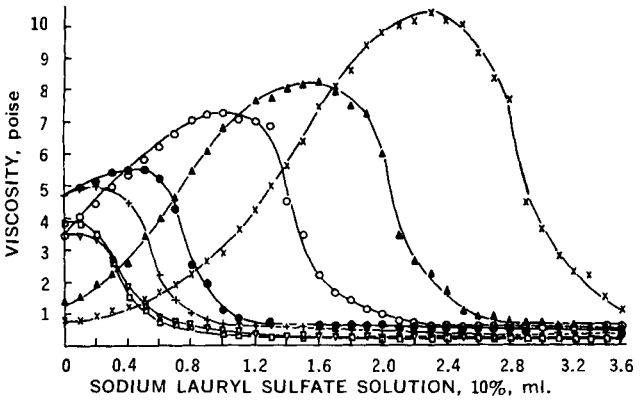

Figure 5—*The increase and/or decrease of viscosity in systems containing 1.4% salicylic acid and varying cetrimide concentrations on addition of 10% sodium lauryl sulfate solution at 25°. The sample weight is 50 g. and the shear rate is 78.56 sec.$^{-1}$. Key: cetrimide concentration (%): ▽, 2.8; □, 3.0; +, 3.5; ●, 4.0; ○, 5.0; ▲, 6.0; and ×, 7.0.*

quantity of anionic surfactant. The major factor which may give rise to the marked increase in viscosity is the neutralization of the free cetrimide molecules by the added anionic surfactant, resulting in a removal of the surfactant molecules from the dispersion medium. The presence of free cetrimide molecules is associated with the viscosity of the system. At the same time, a possible consequence of this neutralization is the approach of the macromolecules closer together, forming a compact mesh and giving rise to a more viscous dispersion.

With further sodium lauryl sulfate addition, however, almost all of the free cetrimide molecules are attracted to the additive and neutralization is at its maximum. In addition, an excess of negatively charged surfactant may result. This process leads to the practically complete withdrawal of free cetrimide molecules from the dispersion medium, thus rendering the system unstable. The interspaces in the mesh structure of the macromolecules are not now occupied by positively charged ions, and this situation contributes to a lowering of the viscosity of the system. The decrease in viscosity continues with further addition of sodium lauryl sulfate solution until the viscosity is too low to be measured accurately.

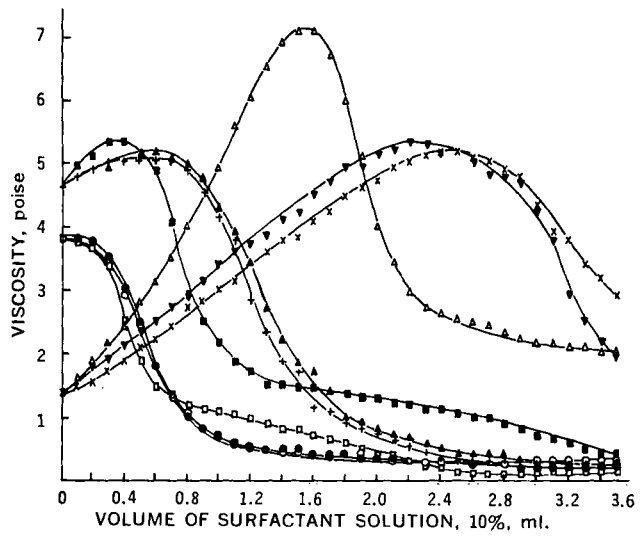

Figure 6—*The increase and/or decrease of viscosity in systems containing 1.4% salicylic acid and varying cetrimide concentrations on addition of solutions of anionic surfactants (10%) at 25°. The sample weight is 50 g. and the shear rate is 78.56 sec.$^{-1}$. Key: cetrimide concentration (10%): for dioctyl sodium sulfosuccinate—□, 3; ■, 4; and △, 6; for sodium alkyl aryl polyether sulfate—●, 3; ▲, 4; and ×, 6; for sodium alkyl aryl polyether sulfonate—○, 3; +, 4; and ▼, 6.*

Table III—Relationship between Amount of Surfactant Required to Decrease Viscosity Initially and Salicylic Acid Concentration of Systems Containing 1.4% Salicylic Acid and Different Cetrimide Quantities[a]

Cetrimide Concentration, %	Weight of Surfactant (g.) which Decreases Viscosity Initially: Sodium Lauryl Sulfate	Sodium Alkyl Aryl Polyether Sulfate	Sodium Alkyl Aryl Polyether Sulfonate	Dioctyl Sodium Sulfosuccinate
3	0.010	0.020	0.020	0.010
4	0.050	0.070	0.080	0.050
5	0.110	0.160	0.150	0.100
6	0.170	0.260	0.230	0.170
7	0.240	0.400	0.330	0.220

[a] Sample weight = 50 g., temperature = 25°, and shear rate = 78.56 sec.$^{-1}$.

No change in viscosity is found when sodium lauryl sulfate solutions are added to cetrimide solutions in the absence of salicylic acid. This fact shows that the viscosity change cannot have been brought about by the simple combination of these surfactants and that the presence of macromolecules is essential.

By fixing the concentration of cetrimide and varying the concentration of salicylic acid, it is possible to produce systems in which the amount of free cetrimide molecules is variable. The lesser the quantity of acid present, the greater is the amount of free cetrimide molecules available. Sodium lauryl sulfate solutions were added to such systems, and the viscosity values obtained are plotted in Fig. 3. The viscosity increases and decreases, except in those systems containing the following concentrations of acid: 1.9, 2.0, and 2.1%. The pattern of viscosity change for systems in which the acid concentration ranges from 1.2 to 1.8% resembles that illustrated in Fig. 1.

Table II shows that as the salicylic acid content is increased, the quantity of anionic surfactant causing an initial viscosity reduction after the attainment of maximum viscosity is decreased. This means that when a smaller number of free cetrimide molecules is available as a consequence of a greater salicylic acid concentration being present, less of the anionic surfactant is required for neutralization.

In those systems (Fig. 3) containing salicylic acid near or in excess of saturation (1.9 or 2.0% and above, respectively), a minimum number of free cetrimide molecules is present in the dispersion, because most of them have interacted with the acid. Hence, on addition of the anionic surfactant, these negatively charged ions are attracted to almost all the available positively charged ions of the cetrimide molecules. This causes the immediate depletion of the free cetrimide molecules from the dispersion medium, the network of macromolecules is weakened and probably disrupted, and the viscosity commences to decrease. Similar results were obtained with the other anionic surfactants investigated (Fig. 4). The acid concentrations used which produced the same effect as did sodium lauryl sulfate are 1.4 and 1.6%.

To obtain different degrees of saturation, salicylic acid concentration is maintained at 1.4% and the cetrimide concentration is varied. An increase in viscosity followed by a decrease is produced in the presence of sodium lauryl sulfate (Fig. 5), except in those dispersions containing 3% or less cetrimide. With a concentration of 1.4% salicylic acid and 3% cetrimide, the system is slightly above saturation, since a few crystals of the acid can be seen after being allowed to rotate in a thermostatically controlled water bath at 25° for 24 hr. Only a reduction in viscosity is produced in dispersions containing 3 and 2.8% cetrimide, in the same manner as in those in which the acid concentration is adequate to saturate or almost saturate the system (Figs. 3 and 4). Sodium alkyl aryl polyether sulfate, dioctyl sodium sulfosuccinate, and sodium alkyl aryl polyether sulfonate exhibit similar behavior (Fig. 6). Cetrimide concentrations of 5 and 7% also demonstrate an increase and decrease in viscosity.

Table III shows that the amount of anionic surfactant added to bring about an initial decrease in viscosity increases with cetrimide concentration. This indicates that when more free cetrimide molecules are present, more anionic surfactant molecules are attracted

to these positively charged ions. These results support the finding expressed in Table II.

The investigation provided further information on the viscous nature of the salicylic acid–cetrimide system and its stability toward surfactants as additives.

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COMMUNICATIONS

Inhibitors of t-RNA *O*-Methyltransferase as Possible Antineoplastic Agents

Keyphrases □ t-RNA *O*-methyltransferase inhibitors—as potential antineoplastic agents □ Antineoplastic agents, potential—inhibitors of t-RNA *O*-methyltransferase

Sir:

In addition to the four main nucleoside constituents (adenosine, cytidine, guanosine, and uridine), *transfer* or *soluble* ribonucleic acids (t-RNA's or s-RNA's) are generally characterized by the presence of a definite amount of methylated nucleosides as minor components (1–10). These methylated nucleosides in t-RNA's are not incorporated as such (11) but are formed at the polynucleotide level by a group of enzymes which catalyze the transfer of methyl groups (methyltransferases, methylases, or transmethylases) from the coenzyme *S*-adenosylmethionine to the t-RNA macromolecules (12–19). The distribution of these methylated units is by no means random and differs in each species (20–22), indicating the existence of certain specificity in the biosynthetic reactions. Viral infection or induction may affect the level of t-RNA methyltransferases (23, 24). It is well known that undermethylated t-RNA's have comparatively inferior aminoacylation activities, codon recognization, and function in protein synthesis (25–29).

It was recently noted that abnormally high levels of methyltransferase enzymes *and* methylase activity, as well as some possible change of specificity of these enzymes, occurred in a variety of neoplastic tissues including virally induced, chemically induced, and spontaneous tumors (30–48). Also reported was the observation that the t-RNA's of many tumors, including both the experimental solid and ascites tumors in animals, as well as human brain tumors, Burkitt lymphoma, glioblastoma, *etc.*, contain highly elevated amounts of methylated—"hypermethylated"—nucleosides (30, 43, 46, 49–52). Since t-RNA's are closely associated with the regulation of protein synthesis at the translation level (26, 53–62) and since alkylating carcinogens were found to alkylate t-RNA to a great extent *in vivo* (43, 63–68) it was postulated that the aberrancy of methyltransferases may be involved in the initiation of tumor induction and neoplasia (32, 66–69). This hypothesis has since received support from other investigators (68–76) and has been considered as one of the most unique and significant findings in cancer research.

Aside from the levels found in neoplastic tissues, larger than normal concentrations of t-RNA methylases were noted in embryonic liver (77) and in chick oviduct (78). Higher t-RNA methylase activity was also observed in fetal brain tissue (79–83). These tissues are characterized by rapid cell division. The activity decreases rapidly in newborn animals after birth (83). It was suggested that the decrease in methyltransferase activity is due to the presence of a methyltransferase inhibitor(s) in adult tissue that is absent in fetal tissue (82, 84). By analogy, it can be postulated that the formation of hypermethylated nucleosides in t-RNA is a result of a lack of methyltransferse inhibitor(s) (85) in the tumor cells. In fact, t-RNA methyltransferase inhibitors, which are found in normal adult rat liver, are virtually absent from the cortex of the highly malignant Walker-256 carcinoma in rats (86). In addition, it was found that an inhibitor prepared from normal adult rat liver had the capacity to inhibit the t-RNA methyltransferase of the Novikoff tumor (82). A search for inhibitors of methyltransferases, therefore, should be of value in cancer chemotherapy, since methylation of t-RNA was shown to be regulated at the enzyme level (87). This is especially true when one considers the possibility that the oncogenic virus might exert its carcinogenicity by introducing a capacity for the synthesis of methyltransferases foreign to the host (15).

Although little is known about actual action of the t-RNA methyltransferase enzymes, information relative to methyltransferase inhibitors may be deduced through an examination of the nature of hypermethylated nucleosides isolated from t-RNA of neoplastic tissues. These nucleosides are composed of the *N*-methylated (*e.g.*, N^6-methyladenosine, N^6-dimethyladenosine, and 1-methylguanosine), the *C*-methylated (*e.g.*, 5-methylcytidine and ribothymine), and the *O*-methylated (*e.g.*, 2′-*O*-methyladenosine, 2′-*O*-methylcytidine, 2′-*O*-methylguanosine, and 2′-*O*-methyluridine) de-