ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, July 1972, p. 23-28

Vol. 2, No. 1
Printed in U.S.A.

Fatty Acids and Derivatives as Antimicrobial Agents

JON J. KABARA, DENNIS M. SWIECZKOWSKI, ANTHONY J. CONLEY, AND JOSEPH P. TRUANT

Department of Microbiology and Department of Osteopathic Medicine, College of Osteopathic Medicine, Michigan State University, East Lansing, Michigan 48823

Received for publication 15 February 1972

The structural relationships of 30 straight-chain fatty acids and derivatives and their bactericidal properties were studied with 8 gram-negative and 12 gram-positive organisms. Chain length, unsaturation (*cis*, *trans*), and functional group were all variables considered in this study. Our data indicate that C_{12} (lauric acid) is the most inhibitory saturated fatty acid against gram-positive organisms. Monoenoic acid ($C_{18:1}$) was more inhibitory than saturated fatty acid, but was less active than dienoic derivatives ($C_{18:2}$). Other unsaturated compounds were less active than $C_{18:2}$. Alcohols and glyceryl esters were active only against gram-positive organisms. In general, esterification of the carboxyl group led to a compound which was less active; monoglycerides were the sole exception. Amine derivatives, contrary to results with fatty acids, esters, and amides, showed activity against both gram-positive and gram-negative organisms.

The antifungal and bactericidal properties of fatty acids are well known (5, 12, 22). In general, fatty acids function as anionic surface agents, and the anionic surfactants are less potent at physiological *p*H values (1, 23). Recently, our interest has focused on the structural and functional role of lipids, especially fatty acids, in microorganisms. In an attempt to correlate fatty acid structure with biological activity at physiological *p*H values, a series of experiments was carried out to obtain semiquantitative information. The information reported herein was obtained with 30 purified compounds which were tested against 12 gram-positive and 8 gram-negative organisms.

MATERIALS AND METHODS

Source of fatty acids. Dodecanedioic acid, trilaurin, cholesteryl laurate, dodecyl-amine HCl, dodecyl acetate, and 1-dodecane-thiol were obtained from Eastman Organic Chemicals, Rochester, N.Y.

Mono- and dilaurin and mono- and dicaprin were kindly submitted to us for study by D. Rebello, Department of Chemical Technology, University of Bombay, Bombay, India.

Except as noted above, the other highly purified fatty acids and related compounds were obtained from Applied Science Laboratories, Inc., State College, Pa.

Preparation of fatty acid solutions. Standard solutions of fatty acids and their derivatives were prepared by first dissolving the weighed compounds in 1 to 2 ml of 95% ethanol and immediately pouring this solution into 200 ml of Trypticase Soy Broth. Those lipid compounds which were insoluble in water and broth precipitated into very small crystals or droplets. The tubes containing the suspensions were warmed to approximately 70 C and were then placed in an ultrasonic tank for further solubilization of the lipid derivatives.

Standard solutions (suspensions) at 1,000 μg/ml were twofold serially diluted with additional broth to a concentration of 125 μg/ml. A 4-ml amount of each dilution was then quickly, and with constant agitation, dispensed into appropriately labeled screw-capped glass tubes (16 by 125 mm). The tubes were sterilized with steam at 15 psi for 15 min. After sterilization, all tubes were cooled and incubated overnight at 35 C to insure sterility.

If a compound was found to have inhibitory activity at concentrations lower than 125 μg/ml, the testing was continued to a final concentration of 1.5 μg/ml for each active compound. All compounds were compared on a molar (micromoles/milliliter) rather than on a mass basis.

Microorganisms. The microorganisms used for our screening studies were those frequently encountered in clinical microbiological specimens. In the following lists of gram-positive and gram-negative organisms used, those followed by a parenthetical number are registered with the American Type Culture Collection; the remainder were isolated at Providence Hospital or Northland Laboratory during 1970. The following gram-positive organisms were tested: *Staphylococcus aureus*, *S. epidermidis*, beta-hemolytic streptococci (group A and non-group A), group D streptococcus, *Bacillus subtilis* (6051), *Sarcina lutea* (9431), *Micrococcus* sp., *Candida albicans*, *Nocardia asteroides* (3308), *Corynebacterium* sp. (10700), and pneumococcus (63001). The gram-negative organisms were *Proteus vulgaris*, *P. mirabilis* (14273), *P. rettgeri* (9250), *Escherichia coli*, *Serratia marcescens* (13880), *Enterobacter-Klebsiella*, *Pseudomonas aeruginosa* (10145), and *Salmonella typhi-*

TABLE 1. *Minimal inhibitory concentrations of 15 fatty acids*[a]

Compound	Pneumococci	Streptococcus group A	Streptococcus beta-hemolytic non-A	Corynebacteria	*Nocardia asteroides*	Micrococci	*Candida*	*S. aureus*	*S. epidermidis*	Streptococcus group D
Caproic	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Caprilic	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Capric	1.45	1.45	2.9	1.45	1.45	2.9	2.9	2.9	2.9	5.8
Lauric	0.062	0.124	0.249	0.124	0.124	0.624	2.49	2.49	2.49	2.49
Myristic	0.218	0.547	2.18	0.437	0.547	0.547	4.37	4.37	2.18	4.37
Myristoleic	0.110	0.110	0.110	0.055	0.110	0.220	0.552	0.441	0.441	0.441
Palmitic	0.48	3.9	3.9	1.9	NI	1.9	NI	NI	3.9	NI
Palmitoleic	0.024	0.098	0.049	0.049	0.049	0.049	0.491	0.983	0.491	0.491
Stearic	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Oleic	NI	1.77	NI	NI	NI	NI	NI	NI	NI	NI
Elaidic	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Linoleic	0.044	0.089	0.089	0.044	0.089	0.089	0.455	NI	NI	NI
Linolenic	0.179	0.35	0.35	0.179	0.448	0.448	NI	1.79	NI	NI
Linolelaidic	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Arachidonic	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI

[a] Results are given in micromoles per milliliter. NI = not inhibitory at the concentrations tested (1.0 mg/ml or 3 to 6.0 μmoles/ml).

murium. Strain differences within a genus were not significant by our mode of testing (*unpublished data*).

One drop (0.04 ± 0.01 ml) of an 18-hr broth culture containing 10^9 to 10^{12} organisms/ml was added to each dilution of the test compound, as well as to a tube of plain broth which served as the positive control. After inoculation, the contents of each tube were well mixed, and the tubes were incubated at 35 C in a 5% carbon dioxide atmosphere.

After 18 hr of incubation, the minimal inhibitory concentration (MIC) of each compound was determined for each microorganism. In our study, the MIC is defined as the lowest concentration of compound at which *no* microscopic evidence of growth was observed when turbidity of the inoculated broth dilutions was compared with control tubes. In those instances in which the test compound itself caused turbidity so that the MIC could not accurately be determined, a sample (0.015 ml) of the well-agitated broths in question was inoculated into a Trypticase Soy Agar plate containing 5% defibrinated sheep blood, incubated at 35 C, and examined after 18 hr for bacteriostatic and bactericidal end points. There usually was only a one-tube difference between the bactericidal and bacteriostatic concentrations.

Control procedures. To test the effects of the sterlization procedure on the stability of the unsaturated fatty acids, a control series was undertaken with 10 mg each of oleic and linoleic acid. Each sample was dissolved in absolute methanol, and the volume was reduced in vacuo to 0.25 ml. A 10-ml amount of sterile broth was added to 0.25 ml of the compound, and appropriate dilutions were made from this stock solution. The activity of these solutions was compared with that of sterilized controls.

To test the effectiveness of a well-known surface-active agent, sodium lauryl sulfate (SDS, Sigma Chemical Co.) was used as a chemical control.

TABLE 2. *MIC ranges for 10 susceptible organisms*[a]

Acid	Range (μmoles/ml)	Max tested (μmoles/ml)
Caproic	NI	10
Caprilic	NI	7.8
Capric	1.45–5.8	5.8
Lauric	0.062–2.49	4.9
Myristic	0.218–4.37	4.37
Myristoleic	0.055–0.552	4.4
Palmitic	0.48–NI	3.9
Palmitoleic	0.024–0.983	3.93
Stearic	NI	3.5
Oleic	1.77–NI	3.54
Elaidic	NI	3.54
Linoleic	0.044–NI	3.56
Linolenic	0.179–NI	3.59
Linolelaidic	NI	3.59
Arachidonic	NI	3.2

[a] The 10 susceptible organisms were *Staphylococcus aureus*, *S. epidermidis*, group D streptococcus, group A streptococcus, beta-hemolytic non-A streptococcus, *Corynebacterium* sp., *Nocardia asteroides*, *Micrococcus* sp., pneumococci, and *Candida albicans*. NI = not inhibitory at the concentrations tested.

Because insolubility of the compound is always a problem in testing lipid material, a control series was undertaken to investigate the effect of particulate suspensions on the bactericidal property. Particularly, the relationship of insolubility of a fatty acid to its biological activity was determined by a series of experiments in which lauric acid was used as the model compound as follows. A suspension of lauric acid, as well as a filtrate of the same suspension, was tested for

TABLE 3. *Minimal inhibitory concentrations of dodecyl derivatives*[a]

Derivative	Pneumococci	Streptococcus group A	Streptococcus beta-hemolytic non-A	Corynebacteria	*Nocardia asteroides*	Micrococci	*Candida*	*S. aureus*	*S. epidermidis*	Streptococcus group D
Lauric acid	0.062	0.124	0.249	0.124	0.124	0.624	2.49	2.49	2.49	2.49
Lauryl alcohol	0.067	0.067	0.271	0.135	0.135	0.135	0.135	0.271	0.135	5.4
Laurylaldehyde	0.136	0.136	0.136	0.136	0.136	0.54	0.136	0.272	0.272	NI
Methyl laurate	—	4.6	NI	4.6	4.6	2.3	4.6	NI	NI	4.6
Dodecanedioic acid	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Lauryl acetate	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Cholesteryl laurate	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Dodecylamine HCl	0.056	0.056	0.112	0.028	0.112	0.112	0.112	0.112	0.112	0.112
Lauryl *N*,*N*-dimethylamide	0.054	0.054	0.109	0.054	0.054	0.109	0.109	0.109	0.109	0.109
1-Dodecanethiol	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI

[a] Results are given in micromoles per milliliter. NI = not inhibitory at the concentrations tested.

antimicrobial activity. Bio Gel and Florisil were mixed with the lauric acid-containing broth to remove particulate matter before it was filtered. Albumin (Miles Pentex, Fatty-Acid Free) to a final concentration of 1.1% was added to a suspension of lauric acid to study the effect of added protein on biological activity.

RESULTS

The data for nine gram-positive organisms and *Candida* are presented in Tables 1, 3, and 5. Tables 2 and 4 give the range of activities as well as the maximal concentration tested; Table 5 presents a comparison of free acids (C_{10} and C_{12}) and their glyceryl derivatives.

Control studies. Both methods of compound addition (pre- or poststerilization) gave identical results except with linolelaidic acid. Repeat tests showed that the results for this fatty acid were variable and were dependent on the time, temperature, and pressure of autoclaving. For this compound, the results from the nonsterilized procedure were used.

SDS was noninhibitory for gram-negative organisms, and was inhibitory at 3.47 μmoles/ml, but not less than 0.347 μmoles/ml, for gram-positive organisms.

The presence of suspended material was necessary because it added to the effectiveness of a compound. Broth with laurate crystals not removed was more inhibitory than filtered broth. For group A streptococci, the results were 0.125 μmole/ml as compared to 1.0 μmole/ml (filtered). When Bio Gel and Florisil were used, the inhibition was reduced to greater than 0.99 μmole/ml. Albumin (0.1%) had no effect on the determination of the MIC under the conditions of our experiment.

TABLE 4. *MIC ranges of dodecyl derivatives for 10 susceptible organisms*[a]

Derivative	Range (μmoles/ml)	Max tested (μmoles/ml)
Lauric acid	0.062–2.49	4.9
Lauryl alcohol	0.067–5.4	5.4
Laurylaldehyde	0.136–NI	5.4
Methyl laurate	2.3–NI	4.6
Dodecanedioic acid	NI	4.3
Lauryl acetate	NI	4.38
Cholesteryl laurate	NI	1.76
Dodecylamine HCl	0.028–0.112	4.5
Lauryl *N*,*N*-dimethylamide	0.054–0.109	4.39
1-Dodecane-thiol	NI	4.94
1-Monocaprin	0.1–2.0	4.04
1,3-Dicaprin	NI	3.3
1-Monolaurin	0.045–NI	3.63
1,3-Dilaurin	NI	2.18
Trilaurin	NI	1.56

[a] The 10 organisms were *Staphylococcus aureus*, *S. epidermidis*, group D strepcococcus, group A streptococcus, beta-hemolytic non-A streptococcus, *Corynebacterium* sp., *Nocardia asteroides*, *Micrococcus* sp., pneumococcus, and *Candida albicans*. NI = not inhibitory at the concentrations tested.

Activity of fatty acids. Microorganisms were arranged by degree of susceptibility.

Considering the saturated acids C_6 to C_{18}, one

TABLE 5. *MIC[a] comparison of free acid form with glyceride form*

Compound	Pneumococci	Streptococcus group A	Streptococcus beta-hemolytic non-A	Corynebacteria	*Nocardia asteroides*	Micrococci	*Candida*	*S. aureus*	*S. epidermidis*	Streptococcus group D
Capric acid...	1.45	1.45	2.9	1.45	1.45	2.9	2.9	2.9	2.9	5.8
1-Monocaprin.....	0.1	0.2	0.2	0.2	0.5	0.1	1.0	1.0	1.0	2.0
1,3-Dicaprin..	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Lauric acid...	0.062	0.124	0.249	0.124	0.124	0.624	2.49	2.49	2.49	2.49
1-Monolaurin.....	0.09	0.045	0.09	0.045	0.09	0.09	0.09	0.09	0.09	NI
1,3-Dilaurin..	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI
Trilaurin.....	NI	NI	NI	NI	NI	NI	NI	NI	NI	NI

[a] Results are given in micromoles per milliliter. NI = no inhibition at the concentrations tested.

can see (Table 1) a measurable change in MIC. The average MIC decreased from a maximum for the C_6 and C_{20} (10 and 3.2 μmoles/ml, respectively) fatty acids to a minimum of 1.16 μmoles/ml (range, 0.062 to 2.49) for C_{12}. The greatest bacteriostatic activity was found for lauric acid (C_{12}).

The addition of a *cis* double bond in the Δ^9 position increased the activity of all fatty acids tested (C_{14}, C_{16}, and C_{18}). The geometric configuration around the double bond is important, because the *trans* isomers were not active. As compared with their saturated counterparts, the $C_{14:1}$, $C_{16:1}$, and $C_{18:2}$ derivatives were more highly active.

Addition of a second double bond further increased the bacteriostatic effect of the (*cis*) C_{18} series. The average MIC of stearic acid was greater than 3.5 μmoles/ml, the highest concentration tested; the MIC of oleic acid was 3.4 μmoles/ml, whereas that of linoleic acid was 0.51 μmole/ml.

Linolenic acid ($C_{18:3}$) was not very active, and therefore the inclusion of the third double bond did not greatly lower fatty acid activity against the organisms tested.

Under our experimental conditions, arachidonic acid ($C_{20:4}$) was not active.

Activity of fatty acid derivatives with a change in functional group. The activity of the compounds tested reflected several important relationships between structure and bacteriostatic action. Since lauric acid was one of the most active acids, analogues with this chain length were studied. Results of this screening process are presented in Table 3.

In comparing the free acid with the alcohol and aldehyde, it can be seen that for most organisms the molar values were closely comparable. The notable exceptions were with the staphylococci and *Candida*, against which the free acid was less inhibitory than the other two. For the micrococci, the alcohol was more inhibitory than the acid or aldehyde.

TABLE 6. *MIC of dodecylamine·HCl for gram-negative organisms*

Organism	MIC (μmoles/ml)
Proteus vulgaris.....	1.10
P. rettgeri.....	0.22
Escherichia coli.....	0.22
Klebsiella-Enterobacter.....	1.10
Serratia marcescens.....	0.22
Salmonella typhimurium.....	0.22

Changing the COOH group to $CONMe_2$ also increased the activity. Reduction of the amide group to give an amine (laurylamine·HCl) yielded a compound which was active against both gram-positive and gram-negative organisms (Table 6).

Although the alcoholic hydroxyl group was particularly active, the substitution of sulfur for the oxygen (SH group) caused loss of compound activity.

Effect of esterified fatty acids. Using lauric acid as a model compound, we examined the effect of esterification with various alcohols. Lauric acid esterified with the monohydric alcohols cholesterol and methanol showed no inhibition. The mono-, di-, and triglyceryl derivatives were also examined. The 1,3-dilaurin and trilaurin derivatives were less active than the fatty acid. The monoglyceride (1-mono-laurin) proved to be more active than the free acid.

A more extensive series of glycerides was studied in another experiment. The results (Table 5) were similar to those of the first trial with laurin derivatives. The C_{10} fatty acid, in agreement with the results of the C_{12} series, was more active when made into a 1-monoglyceride.

DISCUSSION

The literature on the effect of surface-active anionic detergents (fatty acids) dates as far back as the work of Clark (6) reported in 1899. Much of the early literature on this subject can be found in reports by Bayliss (2), Kodicek (18), and Nieman (21). In subsequent years, the antifungal and bactericidal properties of fatty acids have been extensively investigated (5, 12, 22). Other reports point to the inactivation of virus by various soaps (24), as well as to the antitumor activity of fatty acids (25). In the past, certain generalizations were made concerning the activity of fatty acids on cells and microorganisms; however, owing to the impurity of the compounds used, the limited number of organisms tested in the same laboratory, or both, interpretation of structure-function relationships is difficult. Also, it must be kept in mind that considerable difficulty surrounds the testing of lipid materials because of their insolubility. Notwithstanding the above objections, a number of generalities have been made which will be useful.

Of the nine straight-chain, saturated fatty acids tested, lauric acid (C_{12}) had the most bacteriostatic activity on gram-positive organisms. This is in agreement with the results of several other investigators (2, 27). The addition of a *cis* double bond increased the activity of a straight-chain fatty acid, as previously shown (2, 10, 18). The addition of a second double bond further increased the toxicity of the compounds to gram-positive bacteria. A third double bond was not as effective. This does not agree with Kodicek (18), who found the toxicity in increasing order: oleic acid < linoleic < linolenic acid. However, Bayliss (2) and Fuller et al. (10) found, in agreement with us, that the MIC of linoleic acid was somewhat lower than that of linolenic acid.

Hence, although some doubt exists as to whether a third double bond confers more or less activity, there is general agreement foı the following: $C_{18} < C_{18:1} < C_{18:2} > C_{18:3}$. A point to be noted in our study is that only the *cis* forms of unsaturated acids were bacteriostatic. Similar bacteriostatic effects were noted by Kodicek (18), Hettche (14), Chaiz and Baud (4), and Dubos and Davis (8).

The free carboxyl group is necessary for activity, because ester formation generally decreased bactericidal activity of the fatty acid. Reduction of the carboxyl group to the aldehyde or alcohol or the change to an amine or amide group increased bacteriostatic effects. The report of Wyss et al. (27) on antimycotic agents does not support this generalization. They found that fatty acid derivatives (aldehydes, acetate, ethyl ester, amide, or substituted amide), although active, were less active than the corresponding acids. They also found the alcohols to be effective only against more susceptible organisms. That such results are *p*H-dependent can be concluded from the work of Gershon et al. (11).

In the present study, conducted at physiological *p*H values, the order of increasing effectiveness was $COOH < CHO < C - OH, < CONMe_2 < NH_3 + Cl$-.

The amine compound (lauryl amine·HCl), in contrast to other derivatives, was active against both gram-positive and gram-negative organisms. The wide-spectrum activity of amine compounds needs further exploration.

Whereas lauric acid and derivatives have demonstrated high activity against gram-positive organisms, the oxidation of the terminal end of the alkyl chain to form a dicarboxylic acid destroys the activity of lauric acid. Wyss et al. (27) reported a similar finding.

The effect of esterification of lauric and capric acid with glyceryl to form 1-, 1,2,3-, and 1,2,3-glycerides was measured. From Table 5, it can be seen that only monolaurin was more active than the free acid. The antitumor activity of monoglycerides, especially monolaurin (17), suggests that these compounds may affect some fundamental process of cellular growth: Hodes and associates (15, 16) have reported detailed studies of the mode of action of synthetic surfactants on tumor cell membranes. Similar compounds have been found which reduce respiration of ascites cells (20) and their incorporation of ^{32}P (7).

However, since SDS was not as active as lauric acid, the mode of action of the fatty acid cannot wholly be explained in physiochemical terms (surface-tension activity; 9, 19). The explanation probably resides in a more complex mechanism(s). Drug action may involve a change in permeability of the cell wall, although our unpublished data with L-forms suggest interference with cellular metabolism. This could be accomplished by complexing (blocking, removing) critical nutrients or by allowing the diffusion of essential metabolites (21). The interrelationship of fatty acid and carbohydrate metabolism suggests further avenues for investigation (3, 13, 26).

Obviously, many alternative explanations are available. More quantitative data are needed before any hypothesis concerning the mechanism of fatty acid action against microorganisms can be put forward.

LITERATURE CITED

1. Armstrong, W. McD. 1957. Surface active agents and cellular metabolism. I. The effect of cationic detergents on the production of acid and of carbon dioxide by baker's yeast. Arch. Biochem. Biophys. 71:137–147.

2. Bayliss, M. 1936. Effect of the chemical constitution of soaps upon their germicidal properties. J. Bacteriol. **31**: 489–504.
3. Borreback, B., and Q. Spydevold. 1970. Decreased hexokinase activity of the rat epididymal fat pad caused by high level of free fatty acids. A possible factor in the mechanism of dietary adaptation. Biochim. Biophys. Acta **201**:215–225.
4. Chaiz, P., and C. Baud. 1947. Etude de la lyse de glavcoma piriformis (Tetrahymena geleii) par l'acide linoléique et quelques autres acides gras. Arch. Sci. Physiol. **1**:3–10.
5. Chattaway, F. W., C. C. Thompson, and A. J. E. Barlow. 1956. Action of inhibitors on dermatophytes. Biochem. J. **63**:648–656.
6. Clark, J. R. 1899. On the toxic effect of deleterious agents on the germination and development of certain filamentous fungi. Botan. Gaz. **28**:289–327.
7. Creaser, E. H., and P. G. Scholefield. 1960. The influence of dinitrophenol and fatty acids on the P^{32} metabolism of Ehrlich ascites carcinoma cells. Cancer Res. **20**:257–263.
8. Dubos K. J. 1947. The effect of lipids and serum albumin on bacterial growth. J. Exp. Med. **85**:9–22.
9. Frobisher, M. 1926. Relations of surface tension to bacterial phenomena. J. Infect. Dis. **38**:66–91.
10. Fuller, R., and J. H. Moore. 1967. The inhibition of the growth of *Clostridium welchii* by lipids isolated from the contents of the small intestine of the pig. J. Gen. Microbiol. **46**:23–41.
11. Gershon, H., R. L. Rodin, R. Parmegiani, and P. K. Godfrey. 1970. Organic fluorine compounds. IV. Synthesis and antifungal properties of a 2-flouro fatty acid amides. J. Med. Chem. **13**:1237–1239.
12. Glassman, H. N. 1948. Surface active agents and their application in bacteriology. Bacteriol. Rev. **12**:105–148.
13. Hales, C. N. 1966. The glucose-fatty acid cycle and the aetiology of diabetes. Proc. Nutr. Soc. **25**:61–66.
14. Hettche, H. O. 1934. Die Wirkung von Fettsauresalzen auf Eryrozyten und Bakterien. Z. Immunitaetsforsch. **83**: 506–511.
15. Hodes, M. E., C. G. Palmer, and D. Livengood. 1961. The action of synthetic surfactants on membranes of tumor cells. II. Titration experiments. Exp. Cell Res. **24**:298–310.
16. Hodes, M. E., C. G. Palmer, and A. E. Warren. 1960. The effect of surface active agents on the permeability to dye of the plasma membrane of Ehrlich ascites cells. Exp. Cell. Res. **21**:164–169.
17. Kato, A., K. Ando, S. Suzuki, G. Tamura, and K. Arima 1969. Antitumor activity of monoglycerides and other esters of fatty acids. J. Antibiot. (Tokyo) **22**:83–84.
18. Kodicek, E. 1949. The effect of unsaturated fatty acids on gram-positive bacteria. Soc. Exp. Biol. Symp. **3**:217–232.
19. Lamar, R. V. 1911. Chemo-immunological studies on localized infections. I. Action on the pneumococcus and its experimental infections of combined sodium oleate and anti-pneumococcys serum. J. Exp. Med. **13**:1–23.
20. Medes, G. A., J. Thomas, and S. Weinhouse. 1960. Metabolism of neoplastic tissue. XV. Oxidation of exogenous fatty acids in Lettre-Ehrlich ascites tumor cells. J. Nat. Cancer Inst. **24**:1–12.
21. Nieman, C. 1954. Influence of trace amounts of fatty acids on the growth of microorganisms. Bacteriol. Rev. **18**:147–163.
22. Prince, H. N. 1959. Effects of pH on the antifungal activity of undecylenic acid and its calcium salt. J. Bacteriol. **78**: 788–791.
23. Scherff, T. G., and J. C. Peck. 1959. Effects of surface-active agents on carbohydrate metabolism in yeast. Proc. Soc. Exp. Biol. Med. **100**:307–311.
24. Stock, C. C., and T. Francis, Jr. 1943. Additional studies of the inactivation of the virus of epidemic influenza by soaps. **40**:869–875.
26. Weber, G., H. J. H. Convery, M. A. Lea, and N. B. Stamm. 1966. Feedback inhibition of key glycolytic enzymes in liver: action of free fatty acids. Science **154**:1357–1360.
27. Wyss, O., B. J. Ludwig, and R. R. Joiner. 1945. The fungistatic and fungicidal action of fatty acids and related compounds. Arch. Biochem. **7**:415–425.