

April 22, 1941.

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2,238,973

SULPHONAMIDE COMPOUND IN OIL

Filed July 26, 1940

3 Sheets-Sheet 1

EFFECT OF SULFANILAMIDE THERAPY
UPON BETA-HEMOLYTIC STREPTOCOCCAL INFECTIONS OF MICE
WHEN THE COMPOUND IS SUSPENDED IN VARIOUS AMOUNTS OF OLIVE OIL

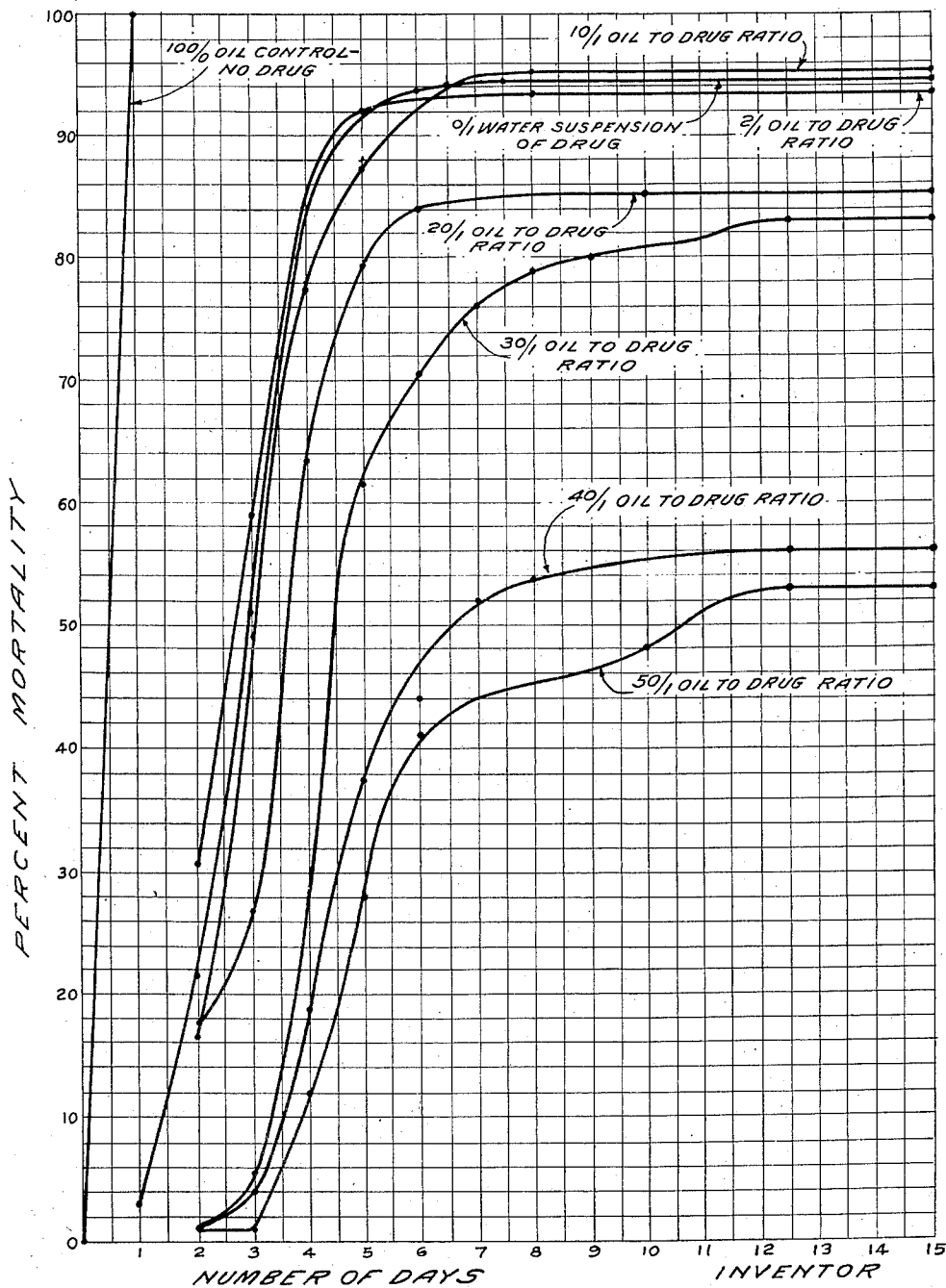


FIG. 1.

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3 Sheets-Sheet 2

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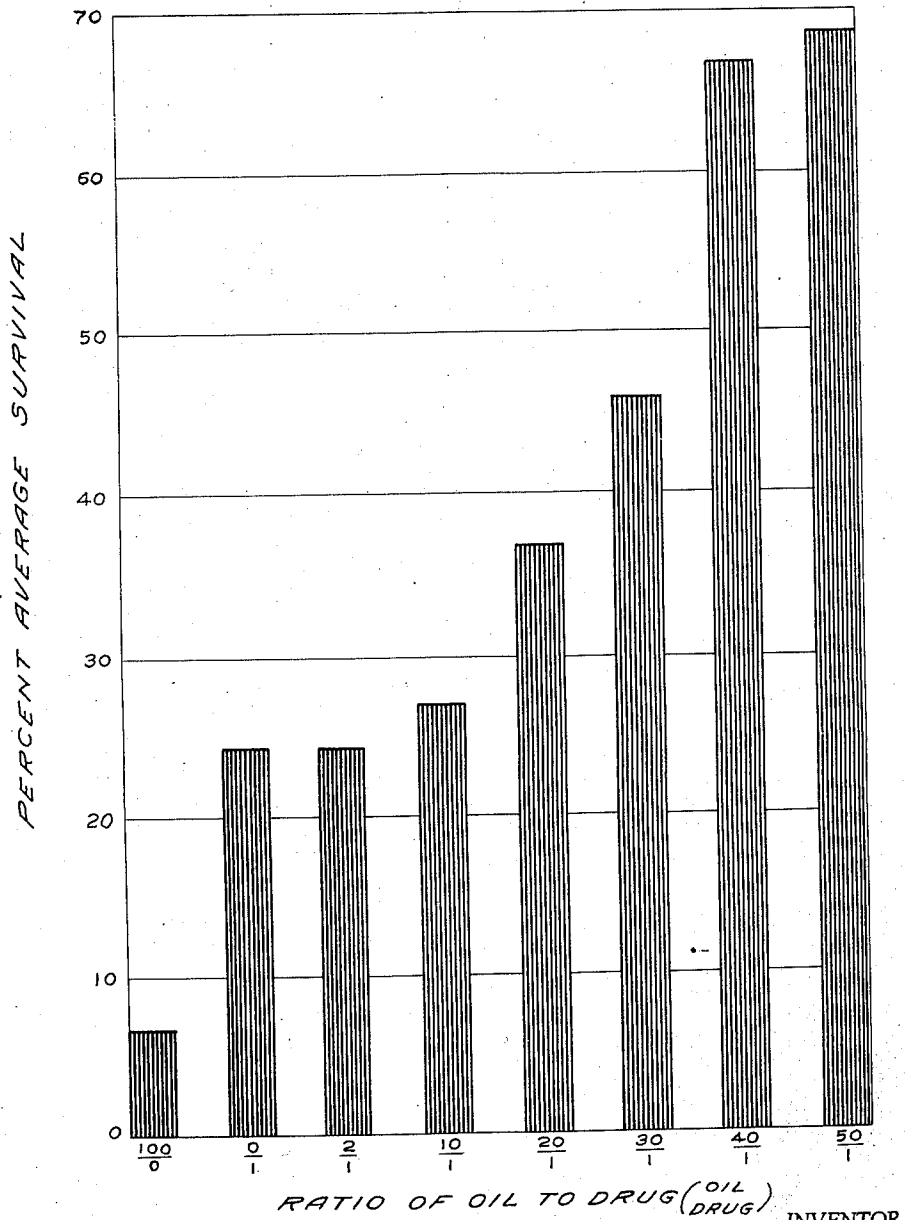


FIG. 2.

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3 Sheets-Sheet 3

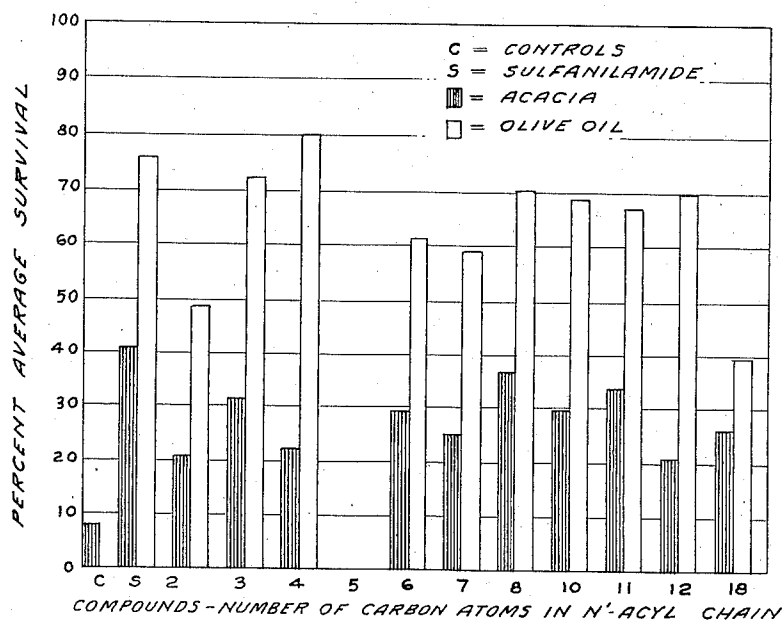


FIG. 3.

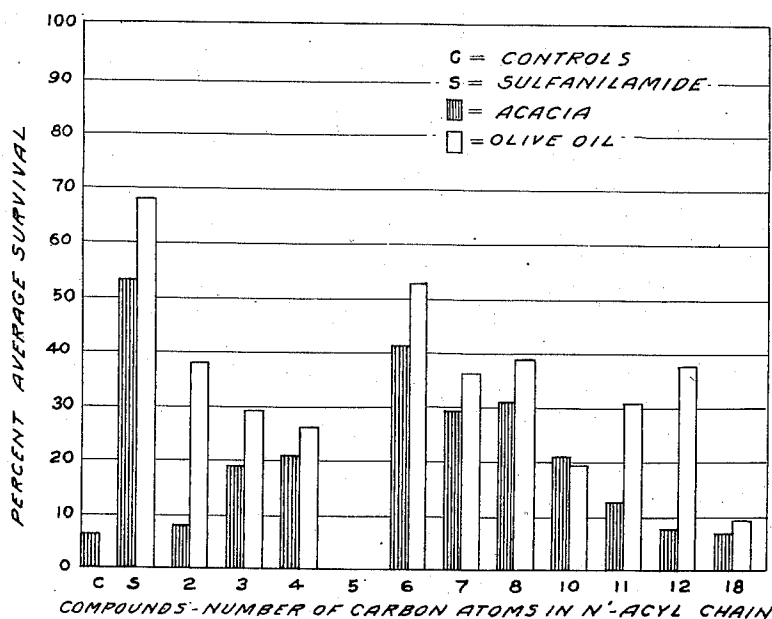


FIG. 4.

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UNITED STATES PATENT OFFICE

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SULPHONAMIDE COMPOUND IN OIL

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Application July 26, 1940, Serial No. 347,638

6 Claims. (Cl. 167-65)

This invention relates to chemotherapy and more particularly to methods and compositions for enhancing the therapeutic properties of sulphonamide compounds particularly for oral administration.

This application is in part a continuation of my application Serial No. 311,227 filed December 28, 1939.

Many sulphonamide compounds are widely used and have been found to be effective in the treatment of a number of infections such as those due to pneumococci, streptococci, meningococci, gonococci, and the like.

According to the present invention I have discovered that the therapeutic effect of the various sulphonamide compounds are enhanced when such compounds are administered orally in a fatty medium provided that certain minimum proportions of the fatty substance are present. When compositions containing sulphonamide compounds and fatty substances are employed in which the ratio of fatty substance to sulphonamide compound is at least 20:1, there is a very marked improvement in the therapeutic effect over the administration of an equivalent quantity of the sulphonamide compound or of compositions containing equivalent quantities of sulphonamide compounds in an aqueous medium. To obtain the best results I prefer to use an oil to drug ratio of from about 40:1 to 50:1.

It is an advantage of the present invention that a wide variety of fatty substances such as solid fats themselves and glyceride oils may be employed to produce the improved results. In fact, any fatty substance that is capable of being assimilated by the body will serve the purpose. The oils and fats are, with few exceptions, esters of the higher members of the fatty acids with the glyceryl radical and hence called glycerides of the fatty acids; therefore not only the oils and fats which are obtained from plant and animal tissue are useful in carrying out the present invention but also the various synthetic glycerides. Among the various fatty substances that have been found to be applicable to the present invention are lard, butter fat, corn oil, coconut oil, cottonseed oil, almond oil, peanut oil, olive oil, sesame oil, some fish oils, suet and synthetic triglycerides such as palmitin, olein, laurein and margarine. These fatty substances while they do have value as foods are considered to be therapeutically inactive.

It is also an advantage of the present invention that the therapeutic properties of all the active sulphonamide compounds are enhanced by administering along with the fatty substance which permits the selection of a particular sulphonamide compound in the treatment of the type of infection for which it has been found to be especially beneficial.

The sulphonamide compounds which have been found to be useful in carrying out the present invention may be represented by the following formula: $(\text{RSO}_2\text{NR}')_n\text{X}$ in which R is an amino or sulphonamido aryl radical, R' is a radical included in the group consisting of thiazoles, pyridines, pyrimidines, hydrogen, alkyl, aryl, or acyl radicals, and X is hydrogen or a metal and n is a small whole number.

Typical sulphonamide compounds useful in the invention include sulphanilamide itself, disulphanilamide, 2-sulphanilamidothiazole, 2-sulphanilamidopyridine, 2-sulphanilamidopyrimidine and the various N¹-acylsulphanilamides such as N¹-butyrylsulphanilamide and N¹-dodecanoylsulphanilamide and the like.

These compositions of the present invention are not to be confused with compositions in which sulphonamide compounds may have been administered in fatty mediums in which the proportion of oil to drug was in the order of the ratio of about 4:1 or about a 20% concentration of the drug in oil. In these prior compositions, which were usually administered by subcutaneous injections, there was no difference in the therapeutic effect produced than that which could have been obtained by administration of equivalent quantities of the sulphonamide compound in an aqueous medium and the oil was used only for convenience, i. e., for the purpose of subcutaneous injection and to lessen chances for irritation due to the solid material at the site of the injection.

The enhanced therapeutic effects of the sulphonamide compounds when administered in a fatty medium have been proven by a number of experiments, the results of which are summarized in the following tables and are illustrated by the drawings in which:

Fig. 1 is a graph which represents the therapeutic effects of sulphanilamide upon beta-hemolytic streptococcal infections of mice when the compound is suspended in various amounts of olive oil, and illustrates the percentage mortality of the infected mice when suspensions of the drug in olive oil were administered in varying proportions;

Fig. 2 is a chart which represents the therapeutic effects of sulphanilamide upon beta-hemolytic streptococcal infections of mice when the compound is suspended in various amounts of olive oil, and illustrates the percentage average survival when suspensions of the drug in olive oil were administered in varying proportions;

Fig. 3 is a chart which represents the comparative therapeutic effects of various sulphonamide compounds in streptococcal infections of mice when the compounds are administered orally in acacia aqueous suspensions and in olive oil suspensions; and

Fig. 4 is a chart which represents comparative

therapeutic effects of various sulphonamide compounds in pneumococcal infections of mice when the compounds are administered orally in aqueous acacia suspensions and in olive oil suspensions.

A number of series of experiments were conducted to determine the comparative therapeutic effect of various combinations of olive oil and sulphanilamide when administered orally upon experimental beta-hemolytic streptococcal infections in mice. In carrying out the experiments, suspensions were prepared of sulphanilamide in olive oil in which the ratio of oil to sulphanilamide were as follows: 50:1, 40:1, 30:1, 20:1, 10:1, 2:1, 0:1 (water suspension of the drug), and 100:0. Each of these suspensions was then tested for its therapeutic effect upon the experimental infections in mice at the same time and in the same experiment and each experiment was repeated a number of times. In each case streptococcus strain C-203 was used which is a strain of beta-hemolytic streptococcus well known in the literature. The virulence of the strain was carefully checked by control animals all of which died. Administration of the drug was oral in each case, the amount being 10 mgs. per dose suspended in the various quantities of olive oil. The treatments were begun immediately after infection and repeated once daily for a total of three treatments. The mice were then observed for 15 days and the average survival was computed on the basis of the survival for any part of the experimental period as well as for the entire experimental period. The total number of animals was about 75 mice in the case of each of the varied proportions of oil and drug and the cumulative results agree in general with the results in the separate experiments done at various times on the smaller group of 25 mice. Data obtained in this fashion is usually considered more accurate than a single experiment performed with 75 mice. The results of the above tests are shown in the following table.

about 5 mice out of 75 will survive when given 10 mg. oral doses of sulphanilamide in water. This shows that a mixture of olive oil and sulphanilamide, with oil to drug ratio being 50:1 is about ten times more effective than equivalent doses of sulphanilamide alone.

These results are graphically illustrated in Figs. 1 and 2. Fig. 2 indicates the percent average survival for the entire period of fifteen days and is based upon the total number of mice used in the experiments. This chart shows that when the ratio of oil to sulphanilamide is only 2:1 or 10:1 there is very little improvement over the test using the drug alone (ratio 0:1). However, when the ratio of oil to drug is 20:1 the percent average survival is about 48% as compared to only about 25% survival when the same quantity of the drug is administered in an aqueous suspension. The chart further shows that the percent average survival increases when the ratio of 30:1 is used over that using 20:1. It is further noted that the maximum percent average survival which is about 69% is obtained when the ratio of 50:1 is used and that this was an improvement of only about 1 or 2% over the 40:1 ratio indicating that while ratios of 50:1 or greater can be used without impairing the therapeutic effect, at the same time the improvement would not be much greater than that obtained using the 40:1 ratio. In Fig. 1 the percent mortality of the mice in the accumulated test is illustrated on the graph. The percent mortality is calculated for each of the days throughout the experiment. Here again there is shown to be very little difference between the therapeutic effect of the drug when administered in the ratio of 2:1 and 10:1 oil to drug and administration of an aqueous suspension or solution, while beginning with the 20:1 ratio there is a definite and marked improvement through each of the ratios up to the 50:1. The graph further shows that the rate of mortality is much greater in the case of the drug in aqueous suspension and also when

TABLE No. 1

The comparative therapeutic effects of various combinations of olive oil and sulphanilamide when administered orally upon experimental beta-hemolytic streptococcal infection of mice

Therapeutic mixture	Ratio oil to sulphanilamide	No. of mice	Dose per os	Deaths in days											Survivals		
				1	2	3	4	5	6	7	8	9	10	11-15	No.	Percent	Percent average
2 gms. S. in 100 cc. oil.....	50:1	75	10 mg. or 0.5 cc.....	---	---	1	8	12	10	2	1	---	2	4	35	46.6	68.5
2 gms. S. in 80% oil in water.....	40:1	75	do.....	---	1	2	11	14	5	6	1	---	---	2	33	44.0	60.0
2 gms. S. in 60% oil in water.....	30:1	75	do.....	---	1	3	20	22	7	4	2	1	---	---	13	17.3	45.9
2 gms. S. in 40% oil in water.....	20:1	75	do.....	---	13	7	27	12	4	---	---	---	1	---	11	13.9	36.8
2 gms. S. in 20% oil in water.....	10:1	79	do.....	---	13	26	22	8	5	---	1	---	---	---	4	5.0	27.0
2 gms. S. in 4% oil in water.....	2:1	75	do.....	---	2	14	22	25	6	---	---	---	---	---	5	6.7	24.1
2 gms. S. in 100 cc. water.....	0:1	75	do.....	---	23	21	19	3	4	---	1	---	---	---	4	5.4	24.3
Olive oil controls.....	1:0	50	0.5 cc.....	50	---	---	---	---	---	---	---	---	---	---	0	0	6.7
Controls.....	---	100	0.....	91	8	1	---	---	---	---	---	---	---	---	0	0	7.3

S.—Sulphanilamide.

It is apparent from an examination of the data in the table that when the oil to drug ratio is 20:1 or greater, there is a marked improvement in the therapeutic effect over that of the aqueous suspensions. The data shows that after mice have been given lethal streptococcal infections about 47 mice out of 75 will survive when 10 mg. oral doses of sulphanilamide suspended in olive oil in the ratio of 50 parts of oil to 1 part of sulphanilamide are administered, whereas only

the ratio of oil to drug is only about 10:1 than when the ratio is 20:1 or greater and further, that at the end of five days substantially all of the animals have died that will die in the fifteen day period when the oil to drug ratio is low, whereas a much smaller number have died at the end of five days when the larger ratios of oil to drug are used. This indicates that the infection is not only cured in a greater number of cases but also that the combination of the

drug with larger quantities of oil are effective in delaying the death of many animals that would have died at an earlier date. The enhanced therapeutic effects obtained in the above experiments are not peculiar to sulphanilamide and olive oil and similar results are obtained with any of the therapeutically active sulphonamide compounds and with other fatty substances.

Experiments were also carried out to determine the comparative therapeutic effect of various sulphonamide compounds in streptococcal infections of mice when the compounds were administered in acacia water suspensions and in olive oil suspensions in which the oil to drug ratio is about 50:1. In each case streptococcus strain C-203 was used which is a strain of beta-hemolytic streptococcus well known in the literature. The virulence of the strain was carefully checked by control animals all of which died. The tests were repeated several times in many instances and the results represent the average percent survival for any part of the experiment as well as the whole. Administration of the drug was oral in each case, the amount being 10 mgs. per dose suspended in $\frac{1}{2}$ cc. of olive oil or acacia solution which represents an oil to drug ratio of between 40:1 and 50:1. The treatments were begun immediately after infection and repeated once daily for a total of three treatments. The mice were then observed for 15 days and the average survival was computed on the basis of the survival for any part of the experimental period as well as for the entire experimental period.

The chart of Fig. 3 summarizes the results obtained by a series of these tests. It is noted that in the case of every sulphonamide compound a marked increase in survival was obtained, in some cases amounting to more than double. While the enhanced therapeutic effect of the administration in the presence of glyceride was notable in every compound, the degree varied

and so long as ratios of at least 20:1 oil to drug are used.

Fig. 4 represents a chart of the results of a similar series of tests with pneumococcal infections in mice. The results are in general the same as in Fig. 3 but are not quite as marked in the case of most of the drugs. The strain used was pneumococcus type 1, strain SV-1, which is well known in the literature and which is a strain that is extremely susceptible to chemotherapeutic action. For this reason the drugs show relatively higher activity than they would in true pneumonias as none of these compounds are considered therapeutically practical for pneumococcal infections. The generally enhanced therapeutic activity in the presence of glycerides is, however, maintained.

The reason why the compositions of the present invention containing glyceride oils and fats with the sulphonamide drugs show such markedly enhanced therapeutic properties is not entirely known and the present invention is not intended to be limited to any particular theory. It was thought that possibly the glycerides might produce increased or accelerated absorption of the drugs when administered orally as a somewhat similar phenomenon has been noticed in connection with the administration of some other drugs with glycerides. Accordingly the factor of absorption was investigated by feeding mice 10 mg. doses of the compounds used in making the experiments shown in the chart of Fig. 3 of the drawings, and blood levels were taken. The results are summarized in the following table from which it will be apparent that with the exception of sulphanilamide itself there is no notable increase in absorption of the various drugs in olive oil as compared to suspensions in acacia. The compounds in these tests were also administered orally in $\frac{1}{2}$ cc. of olive oil or acacia solution.

TABLE 2

The blood concentration in mice obtained after oral administration of a series of N¹-acyl derivatives of sulphanilamide

Compound	Dose	Route	Blood levels in mg. percent hours following administration of the compound						
			1	2	4	8	12	24	
Sulphanilamide.....	Mg. 10	Acacia p. o.	20.0	10.9	5.6	1.0		0	
Do.....	10	Olive oil p. o.	34.0	35.8	16.1	11.1	T	3.8	
N ¹ -acetyl.....	10	Acacia p. o.	9.1	4.0	1.1	T	11.1	FT	
Do.....	10	Olive oil p. o.	8.6	4.5	2.6	0.6		FT	
N ¹ -propionyl.....	10	Acacia p. o.	16.6	10.0	2.9	0.6		0.8	
Do.....	10	Olive oil p. o.	12.5	11.5	7.1	2.7		T	
N ¹ -butyryl.....	10	Acacia p. o.	25.0	18.2	10.5	1.7		T	
Do.....	10	Olive oil p. o.	33.3	16.6	8.7	3.8		T	
N ¹ -hexanoyl.....	10	Acacia p. o.	18.8	16.6	12.5	1.9		0.8	
Do.....	10	Olive oil p. o.	18.0	13.3	10.0	6.2		T	
N ¹ -heptanoyl+	10	Acacia p. o.	29.4	29.4	22.2	9.0		0	
Do.....	10	Olive oil p. o.	28.6	25.4	16.6	9.1		T	
N ¹ -octanoyl+	10	Acacia p. o.	30.0	28.6	16.2	T		0	
Do.....	10	Olive oil p. o.	50.0	43.0	33.3	21.7		FT	
N ¹ -decanoyl+	10	Acacia p. o.	20.0	22.2	16.0	T		0	
Do.....	10	Olive oil p. o.	35.3	30.0	16.0	16.0		FT	
N ¹ -dodecanoyl+	10	Acacia p. o.	11.8	13.8	6.8	T		FT	
Do.....	10	Olive oil p. o.	12.5	8.9	6.7	7.3		FT	

+ Alcohol precipitation method.

p. o., per os.

T, trace.

FT, faint trace.

with various compounds, being particularly noticeable with the N¹-butyrylsulphanilamide.

Tests with other fats show similar results and the enhanced therapeutic effect does not appear to depend on the use of a particular glyceride so long as it is readily assimilable by the animal

From a consideration of the above table it is apparent that while slightly accelerated absorption of the compositions of the present invention may be a minor factor in some cases, it is not a major factor because in the case of most of the drugs where there is no material

difference in absorption there is a very marked difference in therapeutic activity. In evaluating the results of the table which represents tests on a large number of animals, it should be noted that absorption varies quite markedly from animal to animal. The figures in the table are averages but the variation which is normally noted in absorption is such that most of the differences are within the normal experimental error or normal variation. The therapeutic results on the other hand show much less variation so that the figures of the chart set out for Fig. 3 of the drawings are quite accurate.

It is an advantage of the present invention that the amount of glyceride fat or oil used above a certain minimum proportion in which oil to drug ratio is at least 20:1, is not critical and the ratio may be varied from 20:1 up to 50:1 and greater. Still larger amounts of fat do not appear to have any further useful effect. In general it may be stated that the increase in therapeutic activity will vary from drug to drug and it also varies to a lesser extent from glyceride to glyceride, and even from the glyceride in one physical form to the glyceride in a different physical form. In general, the fats which are rapidly assimilable such as butter fats can be used in somewhat smaller amounts than is the case with fats which are very slightly assimilable such as certain very high melting fats. In every case, however, the additions of the glyceride fat or oil enhance the therapeutic activity of the drugs as soon as it is present in substantial amounts. The activity rises to a maximum which will vary with the different drugs and further amounts of a glyceride do not show any particular beneficial action.

It is an advantage that the present invention is not limited to any particular form of composition or method of administration. In many cases it is desirable to keep the drug in suspension as the glyceride medium appears to protect the drug from oxidation or other deterioration, particularly when antioxidants are incorporated in the medium such as, for example, thiourea and substituted thioureas. The compounds may also be administered in the form of oil and water emulsions provided the necessary oil to drug ratios are maintained.

The enhanced therapeutic activity of sul-

phonamide drugs when associated with glycerides may be taken advantage of in either of two ways. The dosage of drug may be maintained constant and an improved therapeutic effect obtained, or a smaller dosage may be used to obtain the same therapeutic effect. Of course, methods combining these two may be used and in general the form in which the compounds of the present invention are utilized will depend on whether in a particular case it is more important to reduce toxic effects incident to large dosages or to obtain even greater therapeutic activity. It is thus an advantage of the present invention that it is very flexible and the treatment calculated to give most effective and safest results in any given case may be chosen.

The enhanced therapeutic effect of the present invention is obtained when the drug is introduced through the alimentary system either by oral or rectal administration. The compositions of the present invention are not, in general, suitable for parenteral administration.

What I claim is:

1. A therapeutic composition for oral administration which comprises a sulphonamide compound having therapeutic properties and a fatty substance of the group consisting of oils, fats, and glycerides contained therein, in which the ratio of the fatty substance to the sulphonamide compound is at least 20:1.
2. A therapeutic composition for oral administration which comprises a sulphonamide compound having therapeutic properties and a fatty substance of the group consisting of oils, fats, and glycerides contained therein, in which the ratio of the fatty substance to the sulphonamide compound is from about 40:1 to 50:1.
3. A composition according to claim 1 in which the sulphonamide compound is sulphanilamide.
4. A composition according to claim 1 in which the sulphonamide compound is an N¹-acylsulphanilamide.
5. A composition according to claim 1 in which the sulphonamide compound is N¹-butyrylsulphanilamide.
6. A composition according to claim 1 in which the sulphonamide compound is N¹-dodecanoylsulphanilamide.

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