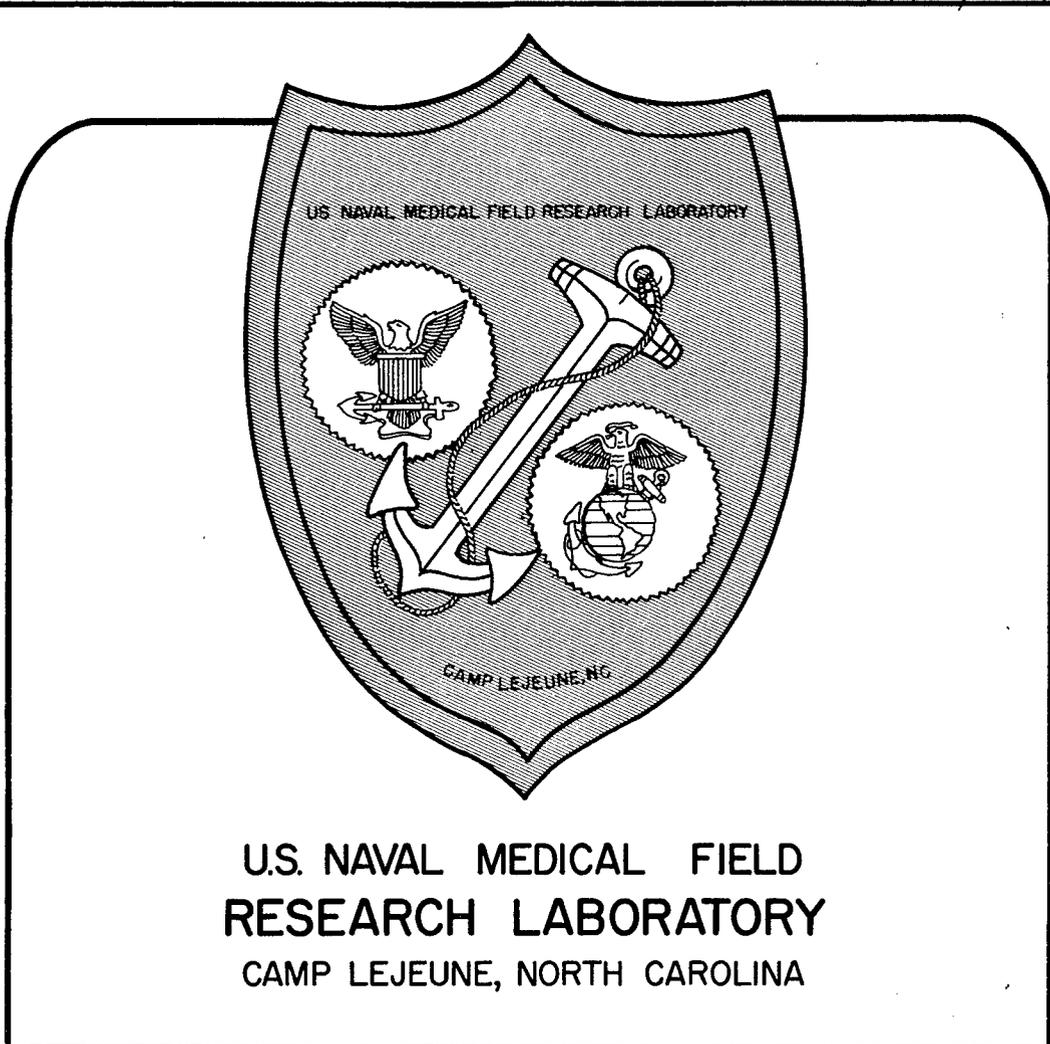


MLC 0213

AD0675813



U.S. NAVAL MEDICAL FIELD
RESEARCH LABORATORY
CAMP LEJEUNE, NORTH CAROLINA

Vol. XVIII, No. 14

August 1968

THE EFFECT OF SELECTED SOFT DRINK MIXES ON THE
GERMICIDAL PROPERTIES OF IODINE TABLETS

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Bureau of Medicine and Surgery, Navy Department
MF12. 524. 007-8007. 1

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20050718027

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MF12.524.007-8007.1

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SUMMARY PAGE

THE PROBLEM

1. To evaluate the effectiveness of tetraglycine hydroperiodide tablets, FSN 6850-985-7166, for purification of contaminated water.
2. To investigate the effects of soft drink mixes on the germicidal effectiveness of the tablets.

FINDINGS

When applied properly, the current method for individual purification with tetraglycine hydroperiodide tablets will provide Marines with purified water. However, the time necessary for these tablets to dissolve and the unsatisfactory packaging may hamper their efficiency and use. Soft drink mixes containing ascorbic acid (Vitamin C) will neutralize elemental iodine and destroy germicidal properties.

RECOMMENDATIONS

1. That improved packaging be developed.
2. That soft drink mixes not containing reducing agents be supplied.
3. That the use of iodine tablets be continued for the present.
4. That consideration be given to developing a disinfectant affording a more rapid dispersion.
5. That a supply of iodine disinfectants prepared and packaged in accordance with the findings of this paper be made available for further testing.

ADMINISTRATIVE INFORMATION

Bureau of Medicine and Surgery, Department of the Navy, Work Unit MF12.524.007-8007, report No. 1. Approved for publication 16 August 1968.

Published by the Naval Medical Field Research Laboratory, Camp Lejeune, North Carolina 28542.

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This restriction will be removed and the report may be released on 30 September 1968.

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ABSTRACT

The germicidal effectiveness of tetraglycine hydroperiodide tablets, FSN 6850-985-7166, was evaluated. The tablets were found to be effective as a germicidal agent. The method used for packaging the tablets makes dispensing them difficult in the field, and permits a reduction in iodine concentration when stored for extended periods.

The effect of soft drink mixes on the germicidal action of iodine tablets was also investigated. It was found that soft drink mixes containing reducing agents (Vitamin C) destroy the germicidal properties of the tablets.

BACKGROUND

During combat operations the availability of purified water is frequently limited. In Vietnam the water available from such sources as shallow wells, streams, and rice paddies is very often contaminated. The waterborne diseases of South Vietnam are typhoid fever, bacillary and amebic dysentery, common diarrhea, cholera, intestinal parasitosis, infectious hepatitis, and leptospirosis.¹

The tetraglycine hydroperiodide tablet, FSN 6850-985-7166, when used properly, has been reported² to provide satisfactory protection against all types of pathogens in nearly all types of waters. Water treated with iodine tablets does have, as would be expected, an iodine taste. This is a small price to pay for their very effective germicidal action, but combat troops often use soft drink mixes to conceal the unpalatable taste of the purified water.

The purpose of this study was (1) to investigate the effectiveness of tetraglycine hydroperiodide tablets for the purification of contaminated water and (2) to determine the results of adding selected soft drink mixes to water in which these tablets are used.

MATERIALS AND METHODS

Effects of Soft Drink Mixes on Elemental Iodine Content

Five hundred milliliters of distilled water were placed in each of four beakers. One tetraglycine hydroperiodide tablet was placed in each beaker and allowed to dissolve. A 2.2 gm¹ Fizzie (R) tablet was added to one beaker, 1.8 gm Fla-Vor-Aid (R) to another, 1.4 gm of Kool-Aid (R) to another, and 39.0 gm of Twist (R) to the final beaker. The manufacturers of these additives as stated on the packages are shown in Table 1. For greater convenience in the tables, they are referred to as mixes Fz, FVA, KA, and Tst, respectively. Only the ascorbic acid, citric acid, sodium bicarbonate, and sodium saccharin were tested for their effects on the germicidal properties of the tablets. Each solution was analyzed for elemental iodine by the solvent extraction method and the starch method,³ and by the method of Johannesson.⁴

Effects of Soft Drink Mixes on Germicidal Action in 10 Minutes

One tetraglycine hydroperiodide tablet was placed in each of four beakers. Each of the foregoing soft drink mixes was added to a separate beaker in the same amounts as used before. Five hundred milliliters of contaminated water were then added to each beaker. The solutions were allowed to stand for 10 minutes. A bacterial colony count was then performed on each solution (Table 2).

Table 1

Brand Name and Manufacturer of Each Soft Drink Mix
Tested for Germicidal Properties of Iodine Tablets

Soft Drink Mix	Brand Name	Manufacturer	Form Supplied
Fz	Fizzie	American Chicle Co. Division, Warner-Lambert Pharmaceutical Company New York, N. Y.	Tablet
FVA	Fla-Vor-Aid	Jel Sert Co. Bellwood, Ill.	Powder
KA	Kool-Aid	General Foods Corp. White Plains, N. Y.	Powder
Tst	Twist	General Foods Corp. White Plains, N. Y.	Powder

Table 2

Bacterial Colony Counts on Solutions Prepared by Adding Soft
Drink Mixes and Tetraglycine Hydroperiodide Simultaneously

Soft Drink Mix	Amount of Mix/500 ml	Bacterial Colony Count/100 ml	
		Before I ₂ Added	After I ₂ Added
Fz	2.2 gm	400 +	400 +
FVA	1.8 gm	400 +	0
KA	1.4 gm	400 +	0
Tst	39.0 gm	400 +	400 +

Effects of Soft Drink Mixes on Germicidal Action in 30 Minutes

One tetraglycine hydroperiodide tablet was placed in each of four beakers. Five hundred milliliters of contaminated water were placed in each beaker. The solutions were allowed to stand for 30 minutes. Soft drink mixes were then added to each beaker as before. A bacterial colony count was then performed on each solution (Table 3).

Table 3

Bacterial Colony Count on Solutions Prepared by Adding the Tetraglycine Hydroperiodide Tablets and 30 Minutes Later Adding the Soft Drink Mixes

Soft Drink Mix	Amount of Mix/500 ml	Bacterial Colony Count/100 ml	
		Before I ₂ Added	After I ₂ Added
Fz	2.2 gm	400 +	0
FVA	1.8 gm	400 +	0
KA	1.4 gm	400 +	0
Tst	39.0 gm	400 +	0

Effects of Selected Soft Drink Components on Free Iodine

Five hundred milliliters of distilled water were placed in each of four beakers. A tetraglycine hydroperiodide tablet was dissolved in each beaker. Ten milligrams of ascorbic acid were added to one beaker, 100 mg of citric acid to another, 100 mg of sodium bicarbonate to another, and 0.20 gm of sodium saccharin to another. Each solution was tested for the presence of free iodine (Table 4).

Preparation of Water Purification Compound

One tetraglycine hydroperiodide tablet was crushed and mixed with 0.15 gm of sodium bicarbonate and 0.15 gm of citric acid. The mixture was placed into a tablet press and 2,000 pounds of pressure were applied for 30 seconds.

Table 4

Test for Elemental Iodine After the Addition of Compounds Found
In Certain Soft Drink Mixes to Water Which Tetraglycine
Hydroperiodide Tablets Had Been Added

Component	Weight of Component	Results of Elemental Iodine Presence In Water Treated With Tetraglycine Hydroperiodide	
		Before Component Added	After Component Added
Ascorbic Acid	0.010 gm	Present	Absent
Citric Acid	0.099 gm	Present	Present
Sodium Bicarbonate	0.10 gm	Present	Present
Sodium Saccharin	0.20 gm	Present	Present*

* Absent after standing 6-8 hours.

Solubility Time

A tetraglycine hydroperiodide tablet was taken from a previously unopened bottle and placed into 500 ml of distilled water with constant stirring. The time required for the tablet to dissolve was recorded. The time required for the compound tablets made by this laboratory to dissolve was determined using the same procedure (Table 5). The color of the tablets was noted and is described in the Results section.

Contact Time for 100% Kill of Pathogens

Five hundred milliliters of contaminated water were placed into each of four beakers. A compound iodine tablet prepared by this laboratory was dissolved in each beaker. One minute after the tablet dissolved, 10 mg of ascorbic acid (Vitamin C) were added to one beaker; 10 mg of ascorbic acid were added to the second beaker 5 minutes after the tablet dissolved; 10 mg of ascorbic acid were added to the third beaker 10 minutes after the tablet dissolved; no ascorbic acid was added to the fourth beaker. A bacterial count was performed on the water before the addition of the tablets, and again immediately after the ascorbic acid was added. The bacterial colony count was performed on the sample with no ascorbic acid added 10 minutes after the iodine tablet had dissolved (Table 6).

Table 5

Length of Time Necessary for Tetraglycine Hydroperiodide
Tablets to Dissolve in Distilled Water at Approximately 20°C

Tablet	Appearance	Time Required for Tablet to Dissolve	
		Minutes	Seconds
1	Light brown	3	10
2	Light brown	3	45
3	Light brown	10	17
4	Light brown	11	2
5	Dark gray	2	25
6	Dark gray	2	28
7	Dark gray	5	32
8	Dark gray	5	38

Note: Tablets 1 and 2 were manufactured in 1965.
Tablets 3 and 4 were manufactured in 1961.
Tablets 5-8 were manufactured in 1966.

Table 6

Time Necessary to Kill All Bacteria Colonies Found
In a Sample of Contaminated Water

Iodine (mg/500 ml H ₂ O)	Weight of Ascorbic Acid (mg)	Contact*		Bacteria Colonies/100 ml	
		Time (min)		Before Iodine Added	After Iodine Added
		Iodine	Ascorbic Acid		
8	10	1	9	830	4
8	10	1	9	830	16
8	0	10	0	830	0
8	10	5	5	830	0
8	10	5	5	830	4
8	10	5	5	830	2
8	10	10	1	830	0
8	10	10	1	830	2
8	0	10	0	830	0
0		0	0	830	-

* Contact time was controlled by adding ascorbic acid to stop the germicidal action.

Elemental Iodine Content of the Tetraglycine Hydroperiodide Tablet

A tetraglycine hydroperiodide tablet was taken from a previously unopened bottle and placed into 500 ml of distilled water. The amount of elemental iodine liberated was measured according to the method of Johannesson.⁴ The instrument used was calibrated by use of a commercially prepared iodine solution (Table 7).

Table 7

Tests for Elemental Iodine After the Addition of Soft Drink Mixes to Solutions Containing Elemental Iodine

Soft Drink Mix	Amount of Mix/500 ml	Elemental Iodine Detected
Fz	2.2 gm	No
FVA	1.8 gm	Yes
KA	1.4 gm	Yes
Tst	39.0 gm	No

Following the observation that the use of certain soft drink mixes reduced the elemental iodine available to disinfect the water, a survey was conducted to determine the water purification practices commonly employed by the combat Marine under field conditions.

A questionnaire was prepared and presented to 139 Marines with a mean tour of 11.9 months in Vietnam.* The greater percentage of these subjects' Vietnam service was in the field and away from base camps. The objective of the questionnaire was to determine the following:

1. The time instructed to wait before drinking the water treated with water purification tablets.
2. Source of water available in Vietnam.
3. Number of purification tablets used in each canteen.

* The authors are indebted to Major James E. Page, USMC, Liaison Officer, Marine Corps Development and Education Command, for assistance in the preparation of this questionnaire.

4. Technique utilized when soft drink mix was used in conjunction with purification tablets.

5. Brand name of soft drink mix used.

6. The reasons for neglecting to use water purification tablets.

7. The percent using a soft drink mix in combination with water purification tablets.

8. The method recommended by Marines for individual purification improvement.

The results of the questionnaire are presented in Table 8, A-G. The questionnaire itself will be found in Appendix A.

Table 8

Results of Questionnaire on Problems of Individual Water Purification

A. Contact time instructed and source of water, with the percent of Marines who used a specific contact time.

Contact Time	Minutes Waited	Percent	Source of Water
Time instructed	<5	14	Streams, wells, rice paddies
Time instructed	5	14	Streams, wells, rice paddies
Time instructed	10	14	Streams, wells, rice paddies
Time instructed	20	12	Streams, wells, rice paddies
Time instructed	30	15	Streams, wells, rice paddies
Failed to state	-	31	Streams, wells, rice paddies

B. Number of purification tablets used in each canteen and percent of Marines using each number.

Number of Iodine Tablets per Canteen			
0	1	2	More than 2
11%	27%	60%	2%

C. Techniques utilized when soft drink mix was used in conjunction with purification tablets and percent of Marines using each technique.

Technique Utilized for Soft Drink Mix	Percent
Iodine and soft drink added simultaneously	20
Used iodine tablets first and waited	49
Used soft drink mix first and added iodine tablets	3
Did not use flavoring agent	14
Failed to state technique	14

D. Brand name of soft drink mix and percent of Marines using each brand.

Brand Name	Percent
Kool Aid	48
Fizzies	17
Funny Face	24
Other brand name	11

E. Reasons given for neglecting to use water purification tablets and percent of Marines giving each reason.

Reasons	Percent
Purification tablets not available	28
Purification tablets taste bad	35
Did not have time	21
Too thirsty to wait for disinfection properties	1
Other reasons, not stated	15

F. Percentage of Marines using a soft drink mix in combination with purification tablets.

Combination Used	Percent
Soft drink mix and purification tablet used	69
No soft drink mix used	29
Failed to answer	2

G. Method recommended by Marines for individual purification improvement.

Method Recommended	Percent
Better instruction on purification methods	2
Add soft drink mix to C-rations	7
Prepare a better tasting purification tablet	25
Better discipline	4
Prepare a method of purification without delay	9
Issue more purification tablets	8
Adopt purification kit similar to Australian	1
Other reasons	4
Failed to state	40

RESULTS AND DISCUSSION

The germicidal properties of the iodine tablets are due to the elemental iodine. Elemental iodine precipitates proteins, the iodine being partly absorbed, partly loosely bound, and partly converted into iodide ions. Since the iodine is loosely bound, it continues to penetrate so that the action extends deeply⁷ into the microorganism.

Our results indicate that soft drink mixes which contain reducing agents (Mixes Fz and Tst) will destroy the germicidal effects of the iodine tablet. This is attributable to the fact that ascorbic acid, a component which was contained in Mixes Fz and Tst of the soft drink mixes, will reduce elemental iodine. This neutralization is indicated by the fact that elemental iodine solutions turn from a brownish color to clear upon the addition of ascorbic acid. Each molecule of ascorbic acid reacts with a molecule of iodine to form dehydro ascorbic acid and two iodide ions, which have little or no germicidal properties.

The ascorbic acid is included in some brands of soft drink mixes to provide the minimum daily requirements of Vitamin C. It is added to the soft drink mixes in the dry state, which is reasonably stable to atmospheric exposure but which deteriorates in aqueous solutions in the presence of air.

Tetraglycine hydroperiodide tablets from freshly opened bottles required from 2 to 10 minutes to dissolve. The color of the tablets when removed from a freshly opened bottle varied from gray to red-brown. The time required to dissolve a gray tablet was about 2-3 minutes. The time required to dissolve a red-brown tablet was approximately 10 minutes. Tablets prepared by this laboratory which contained the same amount of iodine were found to dissolve in 30 seconds or less. The iodine content of the tetraglycine hydroperiodide tablets tested was found to vary from 5.7 mg to 8.8 mg per tablet. The tablets which appeared red-brown in color contained less than 8 mg of elemental iodine per tablet.

Elemental iodine is an effective germicidal agent. Its germicidal action is less dependent on pH, temperature and contact time than are chlorinous nitrogenous disinfectants. Impurities do not impair its effectiveness, and side reactions leading to consumption of the germicide are less marked for iodine than for chlorinous disinfectants.² Elemental iodine is only slightly soluble in water at 20°C; therefore, it cannot be used in water disinfectant tablets.⁵ The iodine may be combined in the form of water-soluble polyiodides. At concentrations of a few parts per million, polyiodides dissociate into elemental iodine.⁵ Morris⁵ reports investigations of several polyiodides. As a result of these tests, he selected tetraglycine hydroperiodide as the compound which best filled the requirements for a good iodine liberating tablet. He concludes that tetraglycine hydroperiodide tablets, when packaged in small amber glass, screw-capped bottles, 50 tablets per bottle, sealed with wax, are stable for extended periods

under conditions of elevated temperatures and high humidity.⁵

The characteristics of a good iodine liberating tablet are as follows:

1. The tablet should dissolve in less than 1 minute at room temperature (26°C).
2. The tablets should liberate at least 8 mg of elemental iodine per tablet.
3. The treated water should be palatable.
4. The tablets should be stable for extended periods of storage.

A convenient method of dispensing the individual tablets should be available. The most significant improvements which might be made in the practice of disinfection of canteen water might include (1) the more rapid release of the disinfectant, (2) improved disinfectant stability, and (3) improved packaging of individual dosages of disinfectant.⁶

The tetraglycine hydroperiodide tablets which we tested were manufactured by Van Brodie 1961-1965. They required from 2 to 10 minutes to dissolve, with constant stirring. Purification tablets prepared by this laboratory, using the standard tetraglycine hydroperiodide tablet, sodium bicarbonate, and citric acid, dissolved in less than 30 seconds.

Protection of iodine tablets from atmospheric humidity during long-time (2 years or more) storage could be improved. Once the wax seal is broken, the tablets are no longer protected from the humidity. Further, when a Marine has wet hands, the current method of packaging makes it difficult to dispense one or two tablets from a bottle without getting water into the bottle, which reduces the germicidal effectiveness of the remaining tablets.

The addition of an iodine tablet, while acting as an effective germicidal agent, does nothing to improve the palatability of the water. The questionnaire disclosed Marines' basic reasons for failure to use purification tablets are: (1) water treated with purification tablets taste badly, (2) purification tablets are not available, and (3) the time element presents a problem. It also revealed that soft drink mixes are used by Marines to make the water more palatable. Soft drink mixes containing reducing agents are not satisfactory for use with iodine tablets. Reducing agents, such as Vitamin C (ascorbic acid) which was contained in two of the mixes tested, immediately destroy all germicidal action of the iodine tablet. All of the elemental iodine present before the addition of the soft drink mix containing ascorbic acid is reduced to iodide, which possesses little or no germicidal action. It was also found that in tests of the individual

components of the four mixes evaluated sodium saccharin acted as a mild reducing agent. Sodium saccharin slowly reduced the germicidal effectiveness of elemental iodine, with complete destruction taking place in about 6 to 8 hours. If any of the germicidal properties of the tetraglycine hydroperiodide tablets are to be realized, the soft drink mixes containing reducing agents should be added at least 30 minutes after the iodine tablet has been added.

One of the major problems of individual water purification is the individual's failure to use the purification tablets. Proper training and troop discipline is considered an important factor in insuring the tablets are used and used correctly.¹

CONCLUSIONS

1. The tetraglycine tablets available for testing required too long to dissolve.
2. The containers currently in use for the tablets are not satisfactory.
3. Soft drink mixes are used in the field to make contaminated water more palatable.
4. Elemental iodine is effective as a germicidal agent, but some of the soft drink mixes used to improve the palatability of contaminated water contain reducing agents, such as ascorbic acid, which destroy the germicidal action of the iodine.

RECOMMENDATIONS

1. To obtain the maximum germicidal effectiveness of the tetraglycine hydroperiodide tablets, a new method of packaging must be used. A possible solution would be to seal the tablets individually in strips of aluminum foil or plastic. This would allow the Marine to tear off one or two tablets and the remaining tablets would still be protected.
2. If the Marines in the field are to be permitted to use soft drink mixes; one which does not contain reducing agents should be supplied.
3. A method of making the use of purification tablets more acceptable to the Marines would be to combine the first two recommendations. A tablet could be prepared which contains tetraglycine hydroperiodide, goes into solution rapidly, has a flavoring agent which does not contain a reducing agent, and is packaged in a manner which will give long-time protection to the tablet and provide for easy dispensing.

4. A method which would employ a disinfecting agent that could be dispersed through the water instantaneously, such as liquid iodine with a buffer, should be developed.

5. Until some method which will automatically disinfect the water when it enters the canteen is discovered, the use of iodine tablets should be continued.

6. Improved annual inspection procedures of tetraglycine hydroperiodide tablets in storage are needed.

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APPENDIX A

WATER PURIFICATION QUESTIONNAIRE

(To be completed by Marines who have served in the Republic of Vietnam)

(Name) (Rank) (Ser #) (Present Organization)

1. What unit or units did you serve with in Vietnam?
2. How many months did you serve in Vietnam?
3. What percentage of your Vietnam service was in the field and away from base camps? _____
4. When operating in the field, from what source did your unit normally obtain the majority of its drinking water?
 - a. Purified water from 5 gallon cans.
 - b. Local water from streams, wells and rice paddies.
5. Did you ever drink local water from?

a. Streams	Yes	No
b. Wells	Yes	No
c. Rice Paddies	Yes	No
6. If you drank local water while in the field, what percentage came from that source? _____
7. Did you use water purification tablets with water obtained from streams, wells and rice paddies?
 - a. Never
 - b. Occasionally
 - c. Frequently
 - d. Always
8. What type of water purification tablets did you use?
 - a. None
 - b. Iodine
 - c. Halazone
 - d. Don't know

9. If you neglected to use water purification tablets in the field on any occasion, please state the reason.

- a. Purification tablets not available
- b. Did not have time
- c. Water purification tablets taste bad
- d. Other (Please state) _____

10. How many water purification tablets did you use in each canteen of local water?

- a. None
- b. One
- c. Two
- d. More than two

11. What period of time were you instructed to wait prior to drinking water treated with water purification tablets? _____

12. When you used water purification tablets, how long did you usually wait prior to drinking the water? _____
(time)

13. Did you ever use a flavoring agent (such as Kool Aid, Fizzies, etc.) when drinking local water?

- a. Never
- b. Occasionally
- c. Frequently
- d. Always

14. If you did use a flavoring agent with local water, what was the brand name?

- a. Kool Aid
- b. Fizzies
- c. Funny Face
- d. Other (Please state) _____

15. When using local water, did you ever use a flavoring agent without purification tablets? Yes _____ No _____

16. Did you ever use a flavoring agent in combination with water purification tablets? Yes _____ No _____

17. If you used a flavoring agent with purification tablets, what technique did you utilize?

- a. Put Iodine and flavoring agent in the canteen simultaneously.
- b. Used Iodine tablets first and waited _____ minutes prior to adding the flavoring agent.
- c. Used flavoring agent first and waited _____ minutes prior to adding Iodine tablets.
- d. Didn't use flavoring agent.

18. If you used a flavoring agent in conjunction with water purification tablets, how long did you wait prior to drinking, after the final ingredient had been added? _____ minutes.

19. When a flavoring agent was used with local water, what was the estimated time before the entire canteen was consumed? _____
(time)

20. Please estimate the average number of canteens of water you drank daily while operating in the field in Vietnam. _____

21. In your opinion, do the Iodine water purification tablets really work?

22. How do you think the water purification system, currently used by the individual Marine, can be improved?

DOCUMENT CONTROL DATA - R & D

(Security classification of title, body of abstract and indexing annotation must be entered when the overall report is classified)

1. ORIGINATING ACTIVITY (Corporate author) Naval Medical Field Research Laboratory Camp Lejeune, N. C. 28542		2a. REPORT SECURITY CLASSIFICATION Unclassified	
2b. GROUP			
3. REPORT TITLE THE EFFECT OF SELECTED SOFT DRINK MIXES ON THE GERMICIDAL PROPERTIES OF IODINE TABLETS			
4. DESCRIPTIVE NOTES (Type of report and inclusive dates) Interim report			
5. AUTHOR(S) (First name, middle initial, last name) Ned S. Hurst, B.S. James S. Bird, LTJG, MSC, USNR			
6. REPORT DATE August 1968	7a. TOTAL NO. OF PAGES 23	7b. NO. OF REFS 7	
8a. CONTRACT OR GRANT NO.		9a. ORIGINATOR'S REPORT NUMBER(S) MF12.524.007-8007.1	
b. PROJECT NO. MF12.524		9b. OTHER REPORT NO(S) (Any other numbers that may be assigned this report) Vol. XVIII, No. 14	
c. Task No. MF12.524.007			
d. Work Unit No. MF12.524.007-8007			
10. DISTRIBUTION STATEMENT This document has been approved for public release and sale; its distribution is unlimited.			
11. SUPPLEMENTARY NOTES		12. SPONSORING MILITARY ACTIVITY U. S. Marine Corps Washington, D. C. 20380	
13. ABSTRACT <p>The germicidal effectiveness of tetraglycine hydroperiodide tablets, FSN 6850-985-7166, was evaluated. The tablets were found to be effective as a germicidal agent. The method used for packaging the tablets makes dispensing them difficult in the field, and permits a reduction in iodine concentration when stored for extended periods. (U)</p> <p>The effect of soft drink mixes on the germicidal action of iodine tablets was also investigated. It was found that soft drink mixes containing reducing agents (Vitamin C) destroy the germicidal properties of the tablets. (U)</p>			

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Interim report, Vol. XVIII, No. 14. Aug 1968 23p
MF12.524.007-8007.1 Unclassified

The germicidal effectiveness of tetraglycine hydroperiodide tablets, FSN 6850-985-7166, was evaluated. The tablets were found to be effective as a germicidal agent. The method used for packaging the tablets makes dispensing them difficult in the field, and permits a reduction in iodine concentration when

stored for extended periods.

The effect of soft drink mixes on the germicidal action of iodine tablets was also investigated. It was found that soft drink mixes containing reducing agents (Vitamin C) destroy the germicidal properties of the tablets.

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