Studies on Ringworm Funguses with Reference to Public Health Problems*

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THE type of skin infection commonly known as ringworm has long been a problem met by the physician treating skin diseases. During recent years this has come to be recognized as a matter of real concern for officers responsible for the administration of certain types of public institutions patronized by large numbers of our population. The high incidence of this type of infection has been repeatedly observed and pointed out. During the past 3 years studies have been carried on at the University of California at Berkeley, by Drs. Robert T. Legge and H. J. Templeton, of the Earnest V. Cowell Memorial Hospital, and the writers of the Department of Botany.[†] Certain aspects of the work have been,⁶ or will be reported elsewhere, while the results of laboratory studies on certain phases of the work are herewith given.

The funguses causing this type of infection are found in the epidermis and the hair growing on infected areas of the body. Skin scales and hair are separated from these lesions and bear the hyphae, Figure I, which are viable and capable of starting a new growth of the fungus given favorable conditions.

Such fungus bearing material is especially apt to be dislodged in dressing rooms and in showers. These infectious materials on the floors of rooms used by numbers of individuals form a ready source of infection for the feet of other persons. The high incidence of this type of infection on the feet of persons using such rooms certainly lends very strong evidence to the support of this as a common method of spread.

Is the fungus material so deposited capable of making actual growth on the floor surface and so multiplying the possible source material for infection? These funguses are well known to be keratin

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loving organisms growing vigorously on materials such as hair, wool, silk, horn and feathers. Data as to their ability to grow on other types of natural materials are very scarce. An effort has been made to collect evidence as to the possibility of growth on materials such as are to be commonly found on the floors of dressing and shower rooms. Repeated examinations have consistently shown the presence of quantities of hair in any cracks in floors, between boards, under lattice work drain boards, and under mats or in cracks in cement.

Microscopic examination of hair from such situations, where there is sufficient moisture, will often show the presence of fungus growth on and in the hair. This type of fungus growth has not been demonstrated to be that of a dermatophyte, due to the great difficult of isolating these comparatively slow growing organisms from such material. Material consisting of hair and other debris from the cracks in floors was brought into the laboratory, moistened with distilled water, and sterilized. Pure cultures of a number of species of ringworm funguses were planted on such material, and all yielded growth, showing that such material, when afforded sufficient moisture, will serve as admirable food material for the growth of these funguses.

The question as to whether these dermatophytes are able to make actual growth on wood as do many other funguses has lacked a satisfactory answer. Specimens of new sound wood of different types,



FIGURE I—Photomicrograph showing mycelium of *Trichophyton interdigitale* imbedded in skin scale

oak, Douglas fir, and pine, were sterilized in flasks after moistening with distilled water, and then inoculated from cultures of a number of species of dermatophytes. These cultures were kept under conditions favorable for possible growth for 6 months' time, but there was no detectable development of fungus in any of the cultures.

Samples of wood were taken from the floor of an outdoor pavilion where the floor boards were somewhat weathered and discolored on the surface due to some years' exposure, but were still apparently sound. These were placed in bottles with distilled water and autoclaved. Blocks of sound new wood were prepared as checks. These were inoculated with fragments from tube cultures of *Trichophyton interdigitale* Priestley, and kept at room temperature. No growth on sound new wood of check. Twelve samples of weathered wood were used. Growth of the fungus developed on three blocks. The growth was very slow in developing and somewhat limited in its extent.

Those failing to show growth were examined by reculturing on agar medium from the inoculum placed on the block, and positive cultures showed that the fungus was viable, but unable to grow except on certain blocks, these apparently affording the food material necessary while others did not. This demonstrated that such old weathered wood may possibly support a growth of this type of fungus.

A point of equal or greater importance to be considered, is whether these funguses may possibly grow on material accumulating on the surface of the floor regardless of the type of floor considered. Surfaces that are subject to frequent wetting tend to accumulate a film or coating of slime. This may in some cases, when not thoroughly cleansed at frequent intervals, show a greenish color due to the growth of algae.

Portions of sound boards, blocks of cement, and bricks that had been thoroughly soaked by continued exposure to wetting for some time, and showed an accumulation of such slime, were taken into the laboratory, placed in culture chambers, and sterilized. The surface was inoculated with a pure culture of *Trichophyton interdigitale*, and they were held at room temperature. A very evident and rapid growth of the fungus resulted (Figs. II and II A). Scrapings of algal growth were made and this material was sterilized and then inoculated with pure cultures. Rapid and abundant growth of the fungus resulted. It is therefore evident that any accumulation of such algal or bacterial slime on the floor or beneath floor mats or drain racks will afford materials for the actual growth and development of this type of fungus, and such growth yields spores readily distributed on any object coming in contact with it. Cracks and fissures in floors would



FIGURE II—Sound wood covered by brown slime, 2 weeks after inoculation with *Trichophyton interdigitale*



FIGURE IIA—Cement block covered with thin greenish slime, 2 weeks after inoculation

often retain sufficient moisture between times of wetting to sustain the growth of the fungus. The cultures shown in Figure II were allowed to dry out in the laboratory and remain dry for 6 months. Subculturing from the surface of these after such drying yielded positive cultures, showing that the fungus may remain viable over rather long periods of ordinary drying. It is, therefore, evident that such growth may remain as a source of dissemination of the fungus over indefinite periods of time.

It appears evident from the data here presented that an important factor in the possible prevention of infection is the elimination of conditions that will permit the possible growth of fungus on floors. Hosing down floors will not be sufficient to remove such material, due to the tenacity with which such materials adhere to the floor. This will be especially true of material under mats and grill work sometimes found on floors and runways. Thorough scrubbing at frequent intervals will be of prime necessity, and possibly the application of disinfectants to the floors.

A series of tests has been made on the action of disinfectants on growths of these funguses in such materials as are found on floors. The materials most likely to serve as food for the growth on floors are: hair, accumulations of slime and algal growth, and skin scales. The preparations used must be such as to be non-irritating to the feet, and the cost of the materials such as to make application practicable.

Zinc chloride solutions are known to have high fungicidal properties, and are used widely as wood preservatives for the prevention of the growth of wood destroying fungus.⁹ Tests on the action of zinc chloride solutions were made as follows: cultures of *Trichophyton interdigitale* were grown on bundles of human hair in culture tubes. When there was vigorous growth on these hair cultures the bundles of hair were laid on blocks of wood that had been placed in large culture dishes and sterilized. Before sterilization the blocks were wet with the zinc chloride solution and enough of the solution provided so that the blocks were about half covered by the solution.

The growing cultures were laid on the surface of the moist blocks, and time allowed for the diffusion of the disinfectant into the hair mat, without immersing it in the solution. This treatment would more nearly approximate the conditions to be met with on a floor surface than would be the case if the growing cultures were simply immersed in the disinfectant solution. Portions of the hair mat were removed at intervals and subcultured for growth of fungus, and results are shown in Table I.

			Time of exposure								
Per cent ZnCl ₂	No. of tests	1 h	our	1 0	lay	2 d	lays	4 d	ays	7 đ	ays
		Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.
13.6	3	3	0	3	0	3	0	1	2	0	3
6.8	13	13	0	12	1	11	2	5	8	0	13
5.0	6	6	0	4	2	4	2	4	2		
5.0	5			4	1	3	2	1	4	0	5
2.0	11	11	0	11	0	11	0	10	1	10	1

TABLE I

ACTION OF ZINC CHLORIDE SOLUTIONS ON CULTURES GROWING ON HUMAN HAIR

The killing action was due to the permeation of the hair mats and strands by the zinc chloride. This penetration was checked by removal of the hair mats after killing and repeatedly washing in distilled water. Tests for chlorides gave positive results after repeated washings, showing that there had been extensive penetration of the hair mats. The results of these tests show a high degree of killing by this agent, in concentrations of 5 per cent or more. The time necessary for killing is rather long, but the high degree of impermeability of the hair, within which much of the fungus material is encased largely accounts for this slow action. It seems that the application of this compound might be advisable to wooden floors, especially in dressing rooms, where the salt would be concentrated in the surface of the wood by evaporation of the water. This residue would then serve to inhibit any fungus growth in organic materials accumulating in cracks in the floor, and apt to become moistened occasionally. Zinc chloride would not seem an advisable disinfectant for the floors of shower rooms, where they are subject to frequent flooding, as it would be washed away before it had time to be effective.

Tests were carried out using some other probable disinfectants, applying them more particularly to accumulations of slime on the surface of wood or blocks of concrete, such as are shown in Figures II and IIA. These slime covered blocks were sterilized in bottles or wide culture dishes, containing small amounts of distilled water, and then inoculated with pure cultures of *Trichophyton interdigitale*. After 2 weeks' time, when a vigorous growth of the fungus had developed on the surface of these blocks, they were transferred aseptically to vessels containing the disinfectant. They were completely immersed in the disinfectant and shaken to insure, as much as possible, thorough contact of the solution with all parts.

After 15 minutes' exposure the blocks were removed to sterile containers and allowed to drain. Subcultures were made at that time in nutrient broth tubes. The blocks were allowed to stand in the containers, and subcultured again after 24 hours. The results of these tests are shown in Table II.

TABLE II

Action of Disinfectants on Fungus Growing on the Surface of Slime Covered Blocks

Material	Time	1 Pe Sodium hy	r cent ypochlorite	1 Per Cus	• cent SO4
Material	Time	Positive	Negative	Positive	Negative
Wood blocks	15 min.	11	3	7	0
Wood blocks	24 hrs.	7	17	7	0
Brick	24 hrs.	0	4		
Concrete	24 hrs.	0	4		

The sodium hypochlorite solution was prepared from standard commercial solutions on the market. The copper sulphate solution was prepared by using the common hydrate $CuSO_4$ ·5H₂O.

The results here shown, although for a small number of tests, indicate that sodium hypochlorite solutions must remain in contact with this material for a considerable period of time to be appreciably effective. The copper sulphate solution is entirely ineffective in the concentration, and for the time intervals used. These results strongly indicate that the probabilities of attaining successful disinfection of such surfaces are slight, unless the disinfectant is to remain on the surface for at least a number of hours. The disinfective action will evidently be in inverse ratio to the amount of organic matter on the surface, and this emphasizes the need for a high degree of cleanliness on such floor surfaces.

A series of tests on the action of sodium hypochlorite on dermatophytes imbedded in skin scales was made. Fragments of skin were collected from cases that on culturing yielded a high percentage of postive cultures of *Trichophyton interdigitale* and *Epidermophyton cruris*. These fragments were soaked in a 1 per cent solution for varying periods of time, washed in sterile water, and planted on nutrient agar. The results are show in Table III.

It is obvious from these results that a high percentage of killing of these funguses is attained only after exposure to the solution for 1 hour or longer. This becomes an important point in connection with the use of such solutions in foot baths. This same sodium hypochlorite solution shows a complete killing action on spore suspensions of these funguses in 5 minutes in dilutions up to 1-5,000. The degree of disinfection attained depends on the readiness with which the solution comes in actual contact with the fungus hyphae. The fungus material imbedded in hairs or in skin scales is encased in material that

DISINFECTANT ACTION OF 1 PER CENT SODIUM HYPOCHLORITE ON SKIN SCALES INFESTED BY DERMATOPHYTES Time of exposure

TABLE III

					Time of	exposure		
Organism	Cor	ntrol	10 m	inutes	30 m	inutes	60 m	inutes
	Plants	Positive	Plants	Positive	Plants	Positive	Plants	Positive
Trichophyton interdigitale	60	33	50	18	50	11	50	3
Epidermophyton cruris	30	14	60	10	10	7	60	0

is very highly impermeable to anything in a watery solution, and this is a fact that must be kept in mind in this work.

Another method of avoiding the possibility of infection from the floor is by the use of bath shoes, to prevent the feet from coming in contact with the floor. Such a procedure is followed in certain institutions, and the evidence to date is such as to lend very strong support to this procedure.^{6b} The enforcement of regulations concerning the wearing of shoes in public institutions adds to the troubles of the officers in charge, and may be difficult in some instances, but the wearing of shoes will eliminate to a very high degree the possibility of the bather coming in contact with the infectious material.

Bath shoes are of various types and materials, such as rubber, paper, leather, and wood. Laboratory tests have shown that the dermatophytes are capable of making growth on leather (Figure III) when it is kept moist, although such growth is exceedingly slow, and many tests have yielded negative results.

Attempts to grow fungus on strips from rubber bathing shoes have shown that it was not able to grow on sponge rubber such as is commonly used in bathing shoes, but one series of tests showed growth on the fabric lining from rubber bath shoes (Figure III). These shoes had been used for some time, and the growth of the fungus might possibly be accounted for by the accumulation of organic matter in the fabric, or by materials with which the fabric was impregnated. This idea is advanced, since we have been unable to demonstrate that these funguses are capable of digesting clean cellulose material. Similar tests made on material from old bathing shoes that were made of sponge rubber alone have yielded only negative results.

THE DISINFECTION OF CLOTHING IN RELATION TO POSSIBLE PREVENTION OF INFECTION

Due to the nature of this disease it is evident that clothing, towels, and bath mats used by individuals suffering from this type of infection may readily bear skin particles permeated by the living hyphae of these funguses. This has been quite generally accepted as a condition of fact; and where such clothing is subject to common laundering there is a strong likelihood that the infectious material may be generally distributed to the articles so handled. This is the basis for the name commonly applied to this type of infection in one country where it is known as "dhobie itch." The name, dhobie, is that applied to the native laundryman.¹

It can be readily demonstrated that these funguses are able to make vigorous growth on wool and silk fabrics, when there is sufficient moisture present, Figure IV. They actually grow in and on the strands of the fabric and digest them, so that eventually there is a breakdown and disappearance of the material of which the fabric was composed. Repeated tests of this type have failed to yield evidence that these funguess are able to attack and digest the cellulose materials of cotton or linen fabrics. Certain statements may be found in the literature ^{3,4} to the effect that they may grow on cotton, but no definite experimental data are given to support this in any report that has come under our survey. Physiological studies show that they require certain types of complex organic nitrogen compounds as a source of nitrogen for their development, and these substances are entirely lacking in materials made up of cellulose.

It is not likely that there would be sufficient moisture in soiled clothing to support an active growth of these funguses in the fabrics under the conditions commonly met with in handling such clothing, although certain cases have been reported where such seemed to be true. There are, however, rather frequently reported cases of infection of the hands of persons engaged in the handling of soiled clothing, as in local laundries.

Studies have been carried on during the past 2 years on the possible disinfecting action of standard laundry and dry cleaning processes on clothing or fabrics bearing viable fungus material.



FIGURE III—Two on left showing growth on fabric lining of rubber bath shoe; center, no growth on sponge rubber; 2 on right, growth on leather from shoe. Age of culture, 6 weeks

FIGURE IV—No discernible growth on cotton, beyond mass of inoculum

TEMPERATURE STUDIES

In an attempt to demonstrate the effectiveness of standard laundry methods in incidental disinfection of articles that carry dermatophytes, the temperature factor is obviously paramount. Verujsky ¹⁰ reported results on thermal death point studies on spore suspensions of *Trichophyton tonsurans* and *Achorion schoenleinii*. He recorded the first killed after 10 minutes at 49° C., the latter not killed in the same time at 50° C. Weidman ¹¹ reported results of some studies on the temperature necessary to kill a number of the species of dermatophytes in culture, and a brief mention of tests on one species when the material was imbedded in skin fragments.

The funguses used in our tests were: Trichophyton interdigitale Priestley, Trichophyton rosaceum Sabouraud, Epidermophyton cruris Castellani, and Microsporon lanosum Sabouraud. These were all grown in pure, bacteria free cultures, and tests were made as follows:

Cultures were grown on agar slants in test tubes until abundant sporulation was obtained. The time necessary for spore formation varies somewhat with different species, but the cultures used varied in age from 15 to 40 days. Spore suspensions were made by introducing into the culture tube a small quantity of sterile distilled water, and then gently working up a spore suspension from the surface of the colony with a soft platinum wire. This spore suspension was then taken off into a sterile tube and agitated to get a homogeneous suspension. Culture tubes were prepared, each containing 10 c.c. of a nutrient solution made up with 1 per cent peptone and 1 per cent glucose.

Five-tenths c.c. of the spore suspension was introduced into each culture tube, and the tubes were then heated for 10 minutes in a water bath which had been brought to the desired temperature. The temperature was read from thermometers held in blank tubes of the same size and containing the same amount of solution. The time interval was reckoned from the time at which the solution within the tube containing the thermometer reached the desired temperature.

At the end of the 10 minutes' exposure the tubes were cooled by immediate immersion in an ice water bath, to insure that the cultures were not exposed to a higher temperature for more than the 10-minute period. Controls were run for each test in tubes inoculated from the same spore suspension and incubated without heating. These cultures were then incubated at room temperature after treatment, and the results read after 14 days. For results see Table IV.

It is a well known fact to students of the dermatophytes that there is a considerable amount of variation in the morphology of these funguses when grown under different cultural conditions. When grown in liquid mediums there is a very considerable development of mycelium without any conidia formation for a period of time. Under certain conditions, especially as the cultures become older, there is a very abundant formation of chlamydospores in the hyphae. Thermal death point studies were made on the four above named species, using

Temper-	T. inter	rdigitale	M. lanosum		E. c	ruris	T. rosaceum		
ature	No. of tests	Positive	No. of tests	Positive	No. of tests	Positive	No. of tests	Positive	
75° C.	14	0	2	0					
70	26	6	12	0			2	0	
65	24	7	20	1	2	0	2	0	
60	24	16	26	5	2	0	8	0	
55	24	22	22	12	2	0	14	0	
50	16	16	18	18	11	0	19	5	
45			14	14	16	9	20	20	
42					12	12			
control	15	15	14	14	5	5	7	7	

TABLE IVConidiospores (Aleuries)

TIME OF EXPOSURE 10 MINUTES

young colonies of vigorously growing mycelium a few days old, before any spores were formed. Another series was run on material from old cultures in which a large percentage of the hyphal cells were rounded and swollen to form chlamydospores, but where conidia formation had been suppressed. The results of these tests made with several dozens of trials for each species showed that the thermal death point for both the young mycelium, and the chlamydospores was consistently a little below that for the conidia shown in Table IV. Since this was found to be true we are reporting here only the results on that phase requiring the higher temperatures to accomplish complete killing.

Cultures Growing on Wool—To each of several 125 c.c. Erlenmeyer flasks 50 c.c. of distilled water was added. To each flask was added 30 small squares (approximately 1 cm. in width) of white Botany flannel. This fabric is a closely woven all wool material. The flasks were sterilized and then inoculated with spore suspensions of the funguses and set aside until profuse growth had taken place, covering all the pieces of the fabric. Microscopic examination showed that the funguses had very completely permeated the fabric, and in some cases the fabric was seen to be digested away to a considerable degree. The squares were then transferred separately to tubes and heated as described above. After heat treatment they were transferred to agar slants and observed for subsequent growth. For results see Table V.

Cultures Growing on Hair—Thick bundles of human hair were placed in test tubes with 3-5 c.c. of distilled water, sterilized, and then inoculated with spore suspensions of the funguses. After sufficient growth had developed (2 to 4 weeks,

Temper-	T. inter	digitale	M. la	nosum	<i>E. c</i>	ruris	T. ros	aceum
ature	No. of tests	Positive						
70° C.	12	0						
65	20	2	2	0				
60	22	6	14	0	18	0	12	0
55	24	13	20	2	21	0	12	0
50	24	23	20	4	21	0	23	0
45	18	18	18	12	22	12	23	22
42	6 ·	6	12	12	4	4	11	11
control	5	5	5	5	5	5	5	5
		С	ULTURES	Growing	on Hair			
65° C.	6	0	6	0				
60	18	0	6	0	24	o	18	0
55	18	0	12	0	24	0	24	1
50	18	11	18	0	24	0	24	7
45	18	18	18	15	24	23	18	18
42			18	18	12	12	6	6
control	2	2	2	2	2	2	2	2

TABLE V Cultures Growing on Wool Time of Exposure 10 Minutes

varying with the species) the entire growth was removed to sterile Petri dishes. The portions of the hair showing abundant growth were selected and separated into small units. These showed abundant growth on and in the hairs when examined under the microscope. Small wisps of infected hair were placed in broth tubes and heated, except for those used as controls. After heating they were cultured on agar slants and observed for further possible growth (see Table V).

Skin Scales Infested with Fungus Material—Since it is obvious that clothing and bath room fabrics used by individuals bearing this type of infection will carry skin fragments infested with these funguses, a series of tests were made to determine the temperature necessary to kill them in this condition. Collections of skin scales were made from patients and tested by laboratory culture. Those showing a high percentage of positive cultures from a number of plants were selected for test materials. The scales were cut into small fragments and subjected to heat treatment for 10 minutes in tubes of distilled water, then taken from the tubes and planted in agar plates. The results from these tests are summarized in Table VI.

IADLE VI	Т	AB	LE	VI
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•						Temp	erature			
Organism	Con	trol	50°	C.	55°	C.	60°	C.	70°	С.
	No. of plants	Posi- tive	No. of plants	Posi- tive	No. of plants	Posi- tive	No. of plants	Posi- tive	No. of plants	Posi- tive
Trichophyton interdigitale	70	50	70	22	40	0	60	0	20	0
Trichophyton rosaceum	20	2	20	2			20	0	20	0

THERMAL DEATH POINT OF TWO SPECIES OF FUNGUS IMBEDDED IN SKIN SCALES. TIME 10 MINUTES

A consideration of the data here listed shows that the dermatophytes are not very highly resistant to heat treatment as applied under moist conditions. The killing temperature is shown to vary somewhat for the different species, *Trichophyton interdigitale* showing the greatest heat resistance, with *Microsporon lanosum* in second place. The effective temperatures here recorded are slightly higher for *Trichophyton interdigitale* than that recorded for complete killing of this species by Weidman,¹¹ but agree with his findings in that this species is more heat resistant than any other used. Our results and those of Weidman do not agree with the statement of Mitchell,⁸ who asserts: "I have been able to culture *Epidermophyton inguinale* from tissue which had been brought to the boiling point in 15 per cent KOH, and obviously ordinary laundering would not kill this organism."

One factor that merits consideration at this point, is the degree of wetting of the material to be treated. In making up spore suspensions one often sees clumping of spores in and about air bubbles in the liquid, and the spores so situated would not be so quickly heated as those more isolated in the liquid. Tests made with *Trichophyton interdigitale* showed that when the spore suspension was shaken with glass beads, decanted, and made up in a more homogeneous suspension, the thermal death point was 5 degrees lower than that shown in our table. Since such a procedure could not be followed in ordinary disinfecting practice it seems better to depend on the figures shown in the table. Practically all laundry treatment of fabrics involves the use of soap, and soap solution serves to lower greatly the surface tension of the bath, and so facilitate complete wetting of all particles.

A number of tests have been made on the possible fungicidal action of soaps. Spore suspensions were made up in 1 per cent soap solutions (on the basis of dry weight), and allowed to stand for periods of 5, 10, and 15 minutes. Subsequent culturing of these spores on a nutrient medium showed no detectable killing or inhibiting action by the soap. Heating of spore suspensions in such soap solutions showed that the temperature necessary for killing action on *Trichophyton interdigitale* is between 60° C. and 70° C. These tests indicate that there is no appreciable fungicidal action exerted on these fungueses by the ordinary soaps that are used in the bath for fabrics, although soaps have been shown to have bactericidal action in such baths, by the work of Elledge and McBride.²

THE EFFICIENCY OF STERILIZATION OF INFECTED ARTICLES BY STANDARD LAUNDRY ROUTINE

Practically all owners of power laundries in the United States are members of the National Association of Laundry Owners. This body functions to promote the establishment of a standard routine procedure most in keeping with the satisfactory functioning of the business, and this standard routine is more or less adhered to by power laundries throughout the country. By reference to their *Manual*⁵ it is found that soiled articles undergo various treatments, which differ primarily according to the fabric, color, weave, and to some extent to the degree to which they are soiled. As the public demands the return of visibly clean and undamaged goods, the laundry finds it necessary to employ the most tempered process that will accomplish this result.

The standard process prescribed for moderately soiled white cotton and linen articles (knitted wear excluded) may be summed up with reference to temperature as follows:

During washing and rinsing such articles are subjected to temperatures of approximately 100° F. (38° C.) for 10 minutes, 120° F. (49° C.) for 5 minutes, and 150° F. (65° C.) for 40 minutes. Soap and soda ash are added to the first two baths. Dirty bundles are subjected to a temperature of approximately 195° F. (90° C.) for 25 minutes. After washing and rinsing the treatment varies with the nature of the article, being either ironed, pressed, or sent to the drying room. Articles ironed or pressed are subjected to a temperature of approximately 320° F. (160° C.) for a short time, while those in the drying room are at 212° F. (100° C.) for approximately 15 minutes.

It is seen from the above that there is little chance of the survival of any of the dermatophytes in view of their heat resistance as shown in the laboratory tests.

Colored cotton articles (except hosiery) are treated as follows:

They are washed for 10 minutes in a bath containing soap and soda ash at 100° F. (38° C.) and this is followed by three 10-minute washings at 120° F.

(49 $^{\circ}$ C.). Rinsing is carried out at this same temperature for about 15 minutes. Following this they are ironed, pressed, or sent to the drying room.

It cannot be stated with certainty, from the viewpoint of the heat resistance of the organisms that this treatment will kill *all* dermatophytes that may be present, although it is obvious that killing would be very nearly complete.

Colored cotton socks, knitted cotton underwear and woolens receive a common type of treatment during the washing process.

The first washing is in soapy water at 100° F. $(38^{\circ}$ C.) for 25 minutes, and this is followed by three rinsings of 5 minutes each at the same temperature. The subsequent treatment again depends on the nature of the fabrics. Since drying at high temperatures causes shrinkage, these articles are not subjected to such high temperatures, nor are they held in the warm bath for such long periods of time as are common cotton articles.

There is no standard procedure for silks, because of the great variation of articles received. Nevertheless, the washing and rinsing temperatures never exceed 100° F. (38° C.), and excessive temperatures for drying or ironing processes are avoided. Silk mixtures and artificial silks are treated as is pure silk.

From the above account it can readily be seen that the killing of the dermatophytes in colored cotton socks, wool, and silk articles is extremely doubtful. It is on such articles, moreover, with the exception of towels, that the occurrence of the infected material is most frequent, because of the nature of the disease.

THE EFFECT OF DRY CLEANING SOLVENTS ON DERMATOPHYTES

Certain types of clothing that are not laundered are subjected to "dry cleaning." Such articles from individuals infected by these funguses may carry infectious material, and recommendations have been made ^{τ} that soaking these articles in dry cleaning solvents would serve to clear them of the infectious material. Such treatment has been recommended for woolen socks to prevent reinfection of cases under treatment. The substances more commonly used for dry cleaning solvents are hydrocarbons, such as gasoline, kerosene, and benzene. Carbon tetrachloride is a component of certain cleaning mixtures.

A series of tests has been made to determine the action of these solvents on four species of dermatophytes. Cultures of the funguses were grown in flasks on fragments of wool fabric and on hair until the mycelium had thoroughly permeated the material. The materials bearing these were removed to dry sterile filter flasks and dried in a 32° C. incubator for 5 days, until they were quite dry. They were then flooded with an excess of the cleaning solvent, and agitated to insure a complete wetting of the surface of the fibers and hyphae of the funguses by the cleaning agent. The agent used in these tests was Cleaners' "naphtha." After the desired time exposure the fluid was. poured off and the wool and hair materials completely dried by passing through the flasks a current of air that had been filtered free of microorganisms. The particles were then removed to culture tubes containing a nutrient medium, and observed for subsequent growth, and their identity was checked. The results of these tests are shown in Table VII.

TABLE VII

RESULTS OF	THE	Exposure	OF	GROWING	Cultures	то	"'Nарнтна"	AND
		SUBS	EQU	JENT RECU	LTURING			

Time in	T. inter	rdigitale	M. la	nosum	Е. с	ruris	T. ros	aceum
solvent	No. of tests	Positive	No. of tests	Positive	No. of tests	Positive	No. of tests	Positive
15 min.	54	54	30	30	30	15	30	30
30 min.	29	29	30	30	30	3	30	30
60 min.	30	30	30	30	60	7	30	10
control	3	3	3	3	4	4	3	3
	1	 		1		l		1

CULTURES G	ROWING C	on Hair
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			obi chilo					
15 min.	30	30	30	30	25	25	30	30
30 min.	30	14	30	28	20	14	30	10
60 min.	30	21	30	10	18	3	30	9
control	4	4	3	3	4	4	4	4

CULTURES GROWING ON WOOL

It is seen from Table VII that there is very little killing action in these species under 1 hour of exposure, and in no case is it complete in 1 hour's time. The percentage of killing is higher with *Epidermophyton cruris* than with the other three species; this species also gives less vigorous growth on hair than the others used.

Tests of the action of naphtha, kerosene, and carbon tetrachloride on spore suspensions showed no appreciable killing action after 5, 10, or 15 minute exposures.

Tests of the possible killing action of some dry cleaning solvents on mycelium of these funguses imbedded in skin scales were made. It is in the skin scales that the fungus material lodged in clothing will

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almost always be found. The action of the solvents on such scales will serve to give us a rather accurate measure of their disinfectant action.

TUDDD ATT	TA	BLE	VIII
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RESULTS OF SOAKING SKIN SCALES BEARING Trichophyton Interdigitale IN SOLVENTS AND SUBSEQUENT CULTURING

Solvent	Control		Time of exposure							
			30 min.		60 min.		90 min.		120 min.	
	No. • of plants	Posi- tive	No. of plants	Posi- tive						
Naphtha	20	7	20	3	20	4			20	4 [.]
Kerosene	30	12					20	. 8		
Carbon tetrachloride	20	7	20	7	20	4			20	5

The results recorded very clearly demonstrate that there is at best a very light disinfectant action by dry cleaning solvents on the ringworm funguess that may be borne on the clothing treated.

There is considerable variation in the procedure followed in the handling of articles to be dry cleaned, depending on the nature of the garment, the degree to which it is soiled, and the equipment of the cleaning establishment. In the better equipped standard establishments the treatment for a great many routine articles is immersion and agitation in the solvent for from 1 to $1\frac{1}{4}$ hours. Where the articles are immersed they must be subsequently dried, and there may be a very appreciable sterilizing action from the drying process, although the temperature is kept in all cases well below 100° C.

SUMMARY

1. Attempts to grow fungus on sound clean wood have yielded only negative results.

2. Fungus may grow readily on floor material that is covered by a coating of slime or algal growth.

3. Results of the application of certain disinfectants to growth on slime covered blocks of floor materials are given.

4. The complete killing of *Trichophyton interdigitale* borne in skin scales, by 1 per cent sodium hypochlorite solution requires a time period of 1 hour or longer.

5. Thermal death point studies on spore suspensions, cultures grown in fabrics, and on material imbedded in skin scales show complete killing of the fungus in 10 minutes' time at 75° C., or lower in some cases.

6. The efficiency of the fungicidal action of standard power laundry practice is shown to vary with the nature of the fabric handled and with the temperature applied to the different materials. The standard practice for white cotton fabrics shows a good margin of safety, while that employed for woolens and colored fabrics is doubtful.

7. The application of standard "dry cleaning" solvents to these funguses, either growing in fabric, or imbedded in skin scales, is shown to have a negligible killing action, in exposures of 1 to 2 hours.

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