an associated unilateral oophorectomy performed. In two others the symptoms arose at a time when irregular menstruation was already present which culminated within 18 months in a complete menopause. Although pain and prolonged cyanosis were dominant symptoms in all cases, only one progressed to digital ulceration within the next five years and in only two cases of these did the symptoms arise *de novo*. In four it was established that mild attacks of pallor or cyanosis had been previously present for many years, and the effect of the menopause in these was to exacerbate a pre-existing mild form of the disease rather than to initiate it.

Mental Stress.—In seven cases the digital symptoms followed a period of prolonged and severe mental stress. In four cases symptoms appeared immediately before or after the death of a mother or husband whom the patients had been nursing, whilst in the three remaining cases they arose during a period of unhappy married life leading to separation or divorce from their husbands. In six of these cases the symptoms arose for the first time within a period of one to four months of either the death of the relative or the date of separation, and in only one case (No. 20) was there any evidence that pre-existing mild attacks of pallor or cyanosis had been present, so that the stress was predominantly an initiating rather than an exacerbating factor in this group of patients.

Discussion

The influence of heredity in Raynaud's disease has previously been commented upon, and it could be seen that in 16 of the 20 cases in which a complete family history was available a history of similar attacks was present in one or both of the parents. It would be reasonable to assume, therefore, that there did exist an inherited predisposition to such attacks in the majority of these patients, although the nature of the inherited defect remains unknown. In the first two groups of 14 cases nine had had symptoms before the event which precipitated the disability, but these had been minimal and had given rise to no disability. In only one of the group associated with mental stress were pre-existing symptoms present. In 10 of the 21 cases, therefore, the effect was to exacerbate a pre-existing mild form of the disease, and in all of these cases (with the exception of Case 7, in which the family history was incomplete) a hereditary history of similar attacks was present. In the remaining 11 cases the symptoms were initiated for the first time by the event, and in seven of these a family history of similar attacks was obtained.

Childbirth and mental stress seem to have been largely disregarded as possible aetiological factors in Raynaud's disease, as, apart from the 137 patients recorded by Allen and Brown (1932b), no large series of cases of this disease have been analysed. It is noteworthy, however, that these authors commented that "in a considerable number of cases the disease appears to be initiated by an operation, childbirth, infection, or the death or illness of a loved one, and these factors could doubtless be elicited in a larger percentage of cases if the patients were questioned specifically regarding this."

In this respect Case 5 in Raynaud's (1862) original thesis was a patient in whom symptoms occurred within a month of the death of the mother, and this was recorded by Hunt (1936) as being the only true example of primary Raynaud's disease in Raynaud's series. Hunt's following case (Mrs. H. G.) was also a patient in whom anxiety of the type described above was present, as she was an individual who was separated from her husband.

The menopause has been stated by Jepson (1951) to have occurred often in the condition he described as "the middle-aged female syndrome." I believe, however, that the use of such a term is not entirely satisfactory, as it is non-specific in character and, according to Jepson's description, covers a number of cases in the age group 35–55 years, in which symptoms were initiated in some instances by the menopause and in others by factors which were not discovered. If all cases of primary Raynaud's disease in this

age group are so classified it is quite probable that a number of cases will be included in which other factors in the aetiology would be disregarded. It should be noted also that, although the menopause is often the cause of symptoms severe enough to give rise to a disability and act as an initiating factor, careful questioning may well reveal that mild attacks had in fact occurred before this but were regarded by the patient as being so mild as to be of no importance.

Whether this latter group should be classified as primary Raynaud's disease or included in secondary Raynaud's disease may be regarded as controversial. Such cases do not exhibit any other signs of disease, and clinical examination fails to reveal any evidence of systemic disease—they therefore fulfil the criteria for a diagnosis of primary Raynaud's disease. To regard these cases as secondary in origin owing to their association with the menopause is no more justifiable than to regard the hereditary group, or those which occur at puberty, as falling into this category. It is believed that these cases should therefore still be included in primary Raynaud's disease.

Summary

Twenty-one cases of primary Raynaud's disease are recorded in which the digital symptoms were initiated, or pre-existing mild attacks were converted into a more severe form, at a time so closely related to childbirth, the menopause, or a period of mental stress as to indicate that these three events should be regarded as aetiological factors in primary Raynaud's disease.

These cases represent 50% of a larger series of 42 cases, in the remainder of which no such aetiological factors could be detected.

REFERENCES

- Allen, E. V., and Brown, G. E. (1932a). *Amer. J. med. Sci.*, **183**, 187.
 — (1932b). *J. Amer. med. Ass.*, **99**, 1472.
 Hunt, J. H. (1936). *Quart. J. Med.*, **29**, 399.
 Jepson, R. P. (1951). *Ann. roy. Coll. Surg. Engl.*, **9**, 35.
 Lewis, T., and Pickering, G. W. (1934). *Clin. Sci.*, **1**, 327.
 Raynaud, A. G. M. (1862). "De l'asphyxie locale et de la gangrène symétrique des extrémités." Thèse de Paris.

A CONTRIBUTION TO THE MYCOLOGY OF THE MOUTH

BY

E. C. FOX, M.D., L.D.S.Ed.

Department of Periodontia, University of Birmingham

AND

G. C. AINSWORTH, Ph.D.

Department of Botany, University of Exeter*

The published records of fungi obtained from the human mouth demonstrate that fungi of many types can be isolated both from apparently normal mouths and from mouths showing various disorders; in some of these diseases fungi have often been regarded as the causal agents. During recent years particular attention has been paid to mycotic infections of the oral cavity as a complication of antibiotic therapy (Cross, 1949; Tomaszewski, 1951a, 1951b, and numerous others).

The literature on the mycology of the mouth, which is both extensive and confused, will be reviewed elsewhere. Here it is only necessary to note that many of the earlier records are unsatisfactory owing to deficiencies in the clinical and mycological evidence, and that only a few attempts have been made to study the mycology of the mouth in relation to a number of fairly common oral pathological conditions.

*Now at the Commonwealth Mycological Institute, Kew, Surrey.

In an effort to supply additional data in this field, isolations of fungi were made from a series of selected cases at Birmingham Dental Hospital during 1952-5 to ascertain whether there were any relationships between fungi and certain well-known pathological conditions. Some of the findings are summarized in this report.

Methods

All the cases studied were subject to very careful clinical examination and detailed case histories were compiled. The isolations were made in the clinic by inoculating slopes of malt extract agar with scrapings taken with a sterile scalpel. Incubation was at 20-25° C., and any fungi which developed were obtained in pure culture for identification. Bacteria and actinomycetes were ignored.

Usually several attempts at isolations were made from each case by taking scrapings from different sites in the oral cavity. For any one case, however, any fungus was recorded once only—for example, as one isolate—irrespective of the number of different sites at which it was found or the number of times it was obtained from one site.

In the control series (groups of schoolchildren, university students, and professional and office workers) some isolations were made as above, in others saliva was sampled by a single swabbing of the tongue, gums, and teeth, and streaking plates or slopes of malt agar with the swab within a few minutes of sampling.

Results

Of the 175 cases examined, 26 that were of miscellaneous conditions are excluded from Tables I and II. These conditions, which showed no distinctive mycological findings, were lichen planus, lupus erythematosus disseminatus, leukoplakia, fungating antrum carcinoma, and dry socket. The remaining 149 cases have been grouped as six reasonably clearly marked clinical conditions—namely, angular cheilosis, gingivostomatitis (comprising aphthous, herpetic, acute ulcerative, mycotic, and various stages of inflammatory periodontal conditions), lingua nigra, lingua geographica, coated tongue, and denture sore mouth—for which the sex and age incidences are summarized in Table I.

TABLE I.—Material

	No. of Cases	Male/Female Ratio	Age	
			Range	Mean
Angular cheilosis ..	24	1.0	16-66	36
Gingivostomatitis ..	36	0.9	4-74	27
Lingua nigra ..	40	2.1	23-68	35
Lingua geographica ..	22	0.27	16-65	37
Coated tongue ..	20	0.43	10-61	38
Denture sore mouth ..	7	0.4	30-78	52
Controls ..	188	1.1	7-60	18

Angular Cheilosis.—This condition is characterized by cracks, fissures, and inflammatory changes at the angles of the mouth, usually accompanied by induration of the edges of the lesion. **Mycology.**—The mycological pattern was very characteristic. Only three of the 24 cases were negative for fungi and 17 of the remaining 21 yielded *Candida albicans*, one of which was in pure culture. Two of the four negative for *C. albicans* gave *C. krusei* and one *C. parapsilosis*—that is, only one case positive for fungi failed to yield a species of *Candida*. Of the other 13 species isolated, *Torulopsis glabrata* was recorded twice and the others were recorded once each. The results show a marked correlation between *C. albicans* and angular cheilosis and support the generally accepted view that *C. albicans* is present as a pathogen.

Gingivostomatitis.—This term is used to describe all inflammatory changes of the gingivae and oral mucosa. As noted above, only aphthous, herpetic, acute ulcerative (Vincent), and mycotic forms and various stages of periodontal disease are included here. **Mycology.**—The general mycological pattern resembled that for angular cheilosis. Although the proportion of cases yielding *C. albicans* was

lower than for angular cheilosis (52.8% as against 70.8%), in 14 of the 19 cases yielding *C. albicans* it was the only isolate. Two other cases gave *C. tropicalis* only. The only other species recorded more than once was *Aspergillus niger* (four times).

Lingua Nigra.—In this tongue condition there is an accumulation of densely matted patches of highly pigmented hypertrophied filiform papillae in front of the circumvallate papillae which may extend into the anterior third. The early literature on this disorder, first described by Rayner in 1835, is summarized by Swinburne (1939), and among other publications on lingua nigra the paper by Kennedy and Howles (1940) and the account by Schaffer (1951-2) may be noted. **Mycology.**—The "random" distribution of isolates from lingua nigra suggested that the isolates were present by chance and bore no causal relation to the clinical condition. This incidence of *C. albicans* was lower than in the control series, but Table II shows the incidence of fungi to have been higher than in either the controls or for angular cheilosis and gingivostomatitis.

TABLE II.—Summary of Isolations From the Controls and From Various Clinical Conditions of the Mouth

Condition	No. of Cases	No. Positive for Fungi	Isolates		
			Total	No. of Species	<i>C. albicans</i>
Angular cheilosis ..	24	21 (87.5%)	36	16	17
Gingivostomatitis ..	36	29 (80.6%)	39	17	19
Lingua nigra ..	40	37 (92.5%)	56	24	9
Lingua geographica ..	22	18 (81.8%)	35	22	8
Coated tongue ..	20	15 (75%)	19	10	4
Denture sore mouth ..	7	6 (85.7%)	8	7	2
Total ..	149	126 (84.5%)	193	A*	59
Controls (normal mouth)	188	124 (66.0%)	162	B*	52

* No figures are shown in A and B as the same species was often recorded for more than one condition. The net total of different species isolated was 54 for A and 37 for B.

Lingua Geographica.—This condition (see Schaffer, 1951-2) is characterized by a circumscribed circinate desquamation of the epithelium of the dorsum and of the edges of the tongue which seems to spread at the periphery whilst it is healing at the centre. It is a chronic condition of unknown aetiology which continuously changes its appearance. **Mycology.**—Table II shows the mycological pattern of the isolates for lingua geographica to be similar to that of lingua nigra. The incidence of *C. albicans* was about "normal" and the incidence of the other species recorded suggested that they were present by chance.

Coated Tongue and Denture Sore Mouth.—Coated tongue is a term applied to a non-specific furred appearance of the tongue. It is a common feature in many diseases of very varied aetiology. Denture sore mouth is a chronic erythema of the denture-bearing area, of the oral mucosa and gingivae which has been attributed to traumatic, allergic, and toxic factors, and to yeasts. **Mycology.**—Fewer fungi were isolated from these two conditions of uncertain aetiology than from the other four, and the more or less random incidence of the isolates suggests that both are non-mycotic.

Discussion and Conclusions

The mycological findings from the clinically abnormal mouth must be considered against a background of the control series summarized in Table II. This Table shows fungi to have been isolated approximately as often from normal mouths (66.0%) as from abnormal (84.5%); the larger number of isolates and species from the abnormal series probably only reflecting the larger number of attempts at isolation. Little evidence was obtained of a characteristic fungus flora of the mouth. Of the 76 different species of fungi recorded, only 15 were common to the two series. Of these, *C. albicans* headed the list with 52 isolates from normal mouths and 59 from the abnormal. Only seven other species were recorded five times or more from abnormal mouths. These were *C. tropicalis* 9 isolates (5 from

the controls), *A. niger* 8 (2), *C. krusei* 7 (1), *T. glabrata* 7 (1), *C. parapsilosis* 6 (5), and *C. zeylanoides* 5 (5). The only species recorded more frequently from the control than from the pathological series was *Aspergillus versicolor* 8 (2), of which six of the isolates occurred in one group of Birmingham schoolchildren. Among all the isolates there was only one of *Absidia ramosa*, an interesting and striking contrast to the frequency of this species among isolates from animal sources (Ainsworth and Austwick, 1955) and from the sputum of agricultural workers.

When the usual significance tests are applied to the material in Table II, it is clear that there is an excess of *C. albicans* isolates in both angular cheilosis and gingivostomatitis (when they are compared with either the other pathological conditions or the controls) which is very unlikely to be due to chance. This is in agreement with the currently accepted view (see Conant *et al.*, 1954), but, bearing in mind the common occurrence of *C. albicans* in the human mouth, predisposing factors must also operate. Many factors may play a part, such as for angular cheilosis lack of vertical dimension with saliva dribbling.

The similarity between the incidence of *C. albicans* in the other conditions (25.8%) and the controls (27.7%) (see Table III) suggests that the control group is reliable and

TABLE III.—Incidence of *C. albicans*

	No. of Cases	<i>C. albicans</i> Isolates	
		No.	%
Angular cheilosis	24	17	70.8 ± 9.3*
Gingivostomatitis	36	19	52.8 ± 8.3*
Other conditions	89	23	25.8 ± 4.6*
Controls	188	52	27.7 ± 3.4*

* Standard error $\sqrt{\frac{pq}{n}}$

that the presence of *C. albicans* seems to be associated with the two specific diseases and not just with any abnormal pathological condition of the mouth. That the isolation of *C. albicans* from the mouth may have no pathological significance hardly needs reiteration, but it should be emphasized that a clinical diagnosis of moniliasis must be confirmed by culture. This is well borne out by the case of a boy aged 19 months who, on examination, showed extensive curd-like whitish plaques on the inner aspects of the upper and lower lip, floor of the mouth, and anterior third of the tongue. The plaques could be peeled off and left raw bleeding surfaces. A clinical diagnosis of oral moniliasis was made, but the only organisms cultured were *Aspergillus fumigatus* and *A. niger*. *C. albicans* is easily cultured and would appear to have been absent in this case. Unfortunately no microscopical examination was made of the fresh clinical material.

The assessment of 37 cases of lingua nigra strongly suggests that this condition is not mycotic. The number of different fungi isolated was greater than for any of the other conditions. No one fungus was isolated with regularity, and the incidence of *C. albicans* was lower than that in the normal mouth. It would appear that the proliferations of the epithelial cells provide a favourable site for the development of common saprophytic fungi. Lingua nigra is thought to be a dyskeratosis. In support of this view further evidence with full case histories will be published elsewhere.

The similar mycological pattern to that of lingua nigra shown by lingua geographica, coated tongue, and denture sore mouth suggests that fungi are not involved in the aetiology of these three conditions. No evidence was obtained for the claim that monilial infections are very often a cause of denture sore mouth. As can be seen from Tables II and III, the incidence of *C. albicans* was rather lower both in denture sore mouth and in coated tongue than in normal mouths, possibly due to the effects of bacterial competition.

It is interesting to record that during this survey no case of mycotic infection of the mouth which could be wholly

attributed to antibiotic treatment was observed. This is in agreement with Heymer and Doeppfmer (1957), who obtained no evidence of a higher incidence of mycotic disease of the oral cavity at the present time than that found by Knighton (1939) before the introduction of antibiotic therapy.

Summary

From the mycological findings in 149 cases of angular cheilosis, gingivostomatitis, lingua nigra, lingua geographica, coated tongue, and denture sore mouth (see Tables II and III) it is concluded that *Candida albicans* seems to play a primary part in the aetiology of angular cheilosis and has some connexion with the aetiology of gingivostomatitis. The diverse fungi associated with the remaining four conditions are of uncertain aetiological significance.

Thanks are due to Dr. C. Rainbow, department of brewing and applied biochemistry, University of Birmingham, for the use of special equipment; to Dr. C. R. Lowe, department of social medicine, University of Birmingham, for advice on statistics; to Mr. D. G. Thomson, Principal School Dental Officer, City of Birmingham, for collecting samples of saliva from schoolchildren; and to the Medical Research Council for training grants which enabled Mrs. June Robson and Mrs. Sheila Wenham to assist with mycological aspects of this investigation.

REFERENCES

- Ainsworth, G. C. and Austwick, P. K. C. (1955). *Trans. Brit. mycol. Soc.*, **38**, 369.
 Conant, N. F., Smith, D. T., Baker, R. D., Callaway, J. L., and Martin, D. S. (1954). *Manual of Clinical Mycology*, 2nd ed., p. 171. Saunders, Philadelphia and London.
 Cross, W. G. (1949). *Brit. med. J.*, **1**, 171.
 Heymer, T., and Doeppfmer, R. (1957). *Arch. klin. exp. Derm.*, **204**, 374.
 Kennedy, C. B., and Howles, J. K. (1940). *Arch. Derm. Syph. (Chicago)*, **42**, 566.
 Knighton, H. T. (1939). *J. dent. Res.*, **18**, 103.
 Schaffer, J. (1951-2). *Oral Surg.*, **4**, 1287; **5**, 87.
 Swinburne, G. (1939). *J. Laryng.*, **54**, 386.
 Tomaszewski, W. (1951a). *Brit. med. J.*, **1**, 388.
 — (1951b). *J. clin. Path.*, **4**, 393.

HAEMOGLOBIN J TRAIT IN TWO INDIAN WOMEN

ASSOCIATED WITH THALASSAEMIA IN ONE

BY

L. D. SANGHVI, Ph.D.

P. K. SUKUMARAN

Human Variation Unit, Indian Cancer Research Centre, Bombay

AND

H. LEHMANN, M.D., Sc.D., M.R.C.P., F.R.I.C.

St. Bartholomew's Hospital, London

Haemoglobin J was first discovered in an American Negro by Thorup, Itano, Wheby, and Leavell (1956). It has since been described in various parts of the world and in people of different racial stock. Raper (1957) found an example in a Gujarati Indian living in Uganda. This paper records the finding of haemoglobin J in two unrelated women belonging to the Gujarati-speaking Lohana. Of particular interest was that one of the two was also carrying a thalassaemia gene, and that it was therefore possible to observe the interaction of thalassaemia and of haemoglobin J.

Techniques

The haematological investigations were carried out by the accepted methods. Osmotic fragility was determined by measuring haemolysis in concentrations varying from 0.1 to 0.7% of Simmell's Tyrode solution, of which 0.4% was