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ORIGINAL ARTICLE

Gastrointestinal Candidiasis in Malnourished Children with Diarrhoea*

by

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Abstract

It is known that the frequency of intestinal Candidiasis in infants and children below 3 years of age is very high, particularly in malnourished children. It has been shown also that Candida can reduce the absorption of sugar, water and electrolytes (Burke et al., 1977; Thelen et al., 1978).

Recently we have studied the frequency of gastrointestinal Candidiasis and the Candida-killing-ability of leucocytes in Indonesian children.

Species of Candida were found significantly more often in throat swabs, gastric, small intestinal contents and stools of malnourished children in comparison with specimens of reasonably well-nourished children.

Furthermore there were marked qualitative differences in the species found in these groups according to their nutritional status.

Studying the Candidacidal activity of leucocytes it shows that in malnourished children the killing-ability is significantly lower than in well-nourished children.

These findings suggest that gastrointestinal Candidiasis may contribute to the pathogenesis of diarrhoea in malnourished children.

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Introduction

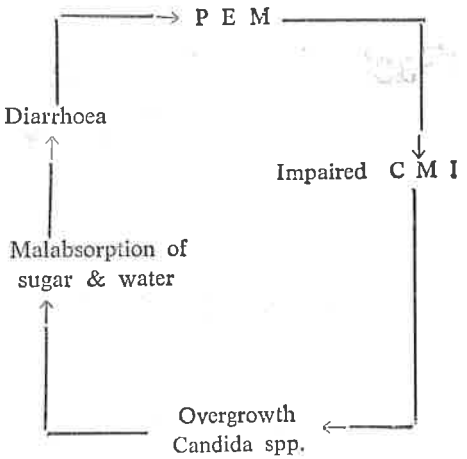
Candida spp. are commonly isolated from humans, particularly in neonates and young infants below 3 years of age (Kozinn et al., 1958; Reyes et al., 1962; Soeprihatin, 1962). They often occur as saprophytes on mucocutaneous surfaces and in the gastrointestinal tract.

Significant infections with Candida spp. appear, however, to be relatively rare except in the presence of predisposing conditions such as malnutrition, impaired cellular mediated immunity or on immunosuppressive therapy, prolonged antibiotic therapy, low birth weight infants, diarrhoea, etc. (Mata et al., 1972; Ono Dewanoto et al., 1968; Suprapti Thaib et al., 1968).

The excessive number of Candida spp. in the gastrointestinal tract may contribute to the production of diarrhoea by their effects on intestinal absorption of sugar (Burke et al., 1977) and fluid and electrolytes (Thelen et al., 1978).

If this condition occurs in malnourished children, the reduction of intake of food and water will increase the degree of malnutrition. Previous studies have shown the ability of such children to secrete intestinal immunoglobulins to be unimpaired (Bell et al., 1976), while depression of cell-mediated immunity in vivo and in vitro had been reported in malnutrition (Geefhuysen et al., 1971; Chandra, 1972; Ferguson et al., 1974).

This condition then will cause the increase of the numbers of Candida spp. in the gastrointestinal tract, so that it became a vicious cycle as shown in figure 1.



The purpose of this paper is to present some results of studies in the relationship between malnutrition, gastrointestinal candidiasis and diarrhoeal diseases in Indonesian children.

Investigations

Several investigations regarding intestinal candidiasis in children with malnutrition and diarrhoea have been done. They consist of:

1. Studying the duodenal and gastric content of 21 malnourished Indonesian children with diarrhoea.
2. Studying the oropharyngeal microflora of 28 malnourished Indonesian children.
3. Isolation of Candida spp. from the gastrointestinal tract in malnourished children.
4. Investigating the frequency of intestinal Candidiasis of infants and children with diarrhoeal diseases.

5. The use of a simple duodenal capsule to study upper intestinal microflora.
6. Studying the Candida-killing-factor in malnourished patients.

Results

The results of these studies are summarized in Table 2 to 12.

1. Microbial contamination of the gut: another feature of malnutrition (M. Gracey, Suharyono, Sunoto and Delys E. Stone, 1973).

In a study of microflora of the stomach and upper small intestine in 21 malnourished Indonesian children with diarrhoea and compared with that found in 21 Australian children of caucasian descent revealed significant overgrowth of Candida spp. The difference between the 2 groups was statistically significant ($p < 0.025$).

In 8 (40%) out of 20 malnourished children the number of isolation was more than 10^4 per ml (Table 3).

2. Oropharyngeal microflora in well-nourished and malnourished children (M. Gracey, Delys E. Stone, Suharyono and Sunoto, 1973).

A study of 28 Indonesian children with protein energy malnutrition and chronic or recurrent alimentary tract infection compared with 42 well-nourished Australian children of Caucasian descent as controls, revealed that in the malnourished group higher isolation of Candida spp. was found (36% versus 7%).

3. Isolation of Candida spp. from the gastrointestinal tract in malnourished children (M. Gracey. Delys E. Stone, Suharyono and Sunoto, 1974).

Gastrointestinal Candidiasis was examined in 2 groups of malnourished children and compared with that in a group of relatively well-nourished children. There were 30 Australian aboriginal children treated because of diarrhoea and malnutrition which was endemic in this group. Similarly, the 27 Indonesian children were suffering from protein energy malnutrition and chronic or recurrent gastrointestinal infections and infestations.

In each of the three groups, antibiotic therapy was used in several patients. This did not significantly influence the rate of isolation of Candida spp. from gastric or small-intestinal contents (Table 5) and comparison of the results in the three groups is, therefore made, using total results.

Nine of the Caucasian children had significant numbers of Candida spp. in their gastric juice (10^4 to 10^7 per ml). Large numbers of Candida spp. (up to 10^9 per ml) were found in gastric aspirates from twenty-one of the aboriginal children and nineteen of the Indonesians. In each instance the rate of isolation was very significantly increased in comparison with the Caucasians (Table 6).

TABLE 1 : *Classification of the nutritional state as suggested by the Wellcome Working Party (1970). Weight expressed as a percentage of standard weight for age (SWFA) compared with the 50th centile of the Harvard Standard*

Nutritional Status	Percentage of SWFA	E d e m a
Normal	> 80	N o
Underweight	60 — 80	N o
Kwashiorkor	60 — 80	Y e s
Marasmus	< 60	N o
Marasmic - Kwashiorkor	< 60	Y e s

TABLE 2 : *Small intestinal microflora in 20 malnourished Indonesian children compared to 21 Australian children*

	Gram-positive Bacteria	Candida	Entero-bacteria	Faecal bacteria	Total Aerobes	Anae-robes	Total Organisms
Indonesian patients							
L_1	3.8	0.7	3.7	0.6	4.6	0.9	4.6
Mean	4.8	2.4	4.5	1.4	5.5	2.0	5.5
L_2	5.8	4.1	5.3	2.2	6.4	3.1	6.4
Range*	0-1 x 10^8	0-2 x 10^6	0-1.3x10^9	0-4x10^6	0-1.3x10^9	0-1.3x10^8	0-1.3x10^9
Controls							
L_1	1.7	0.1	0.5	0.1	2.2	1.9	2.2
Mean	2.3	0.5	1.1	0.6	2.8	0.1	2.8
L_2	2.9	0.9	1.7	1.1	3.4	0.3	3.4
Range*	0-4 x 10^4	0-1.3x10^3	0-3.2x10^4	0-2x10^3	0-5x10^4	0-7.9x10^2	0-5 x 10^4
P	< 0.005	< 0.025	< 0.0005	0.3<p0.35	< 0.0025	< 0.05	< 0.0025

Results indicate population on various species and total flora expressed as the $\log_{10}$ of the mean viable colony count per millilitter of specimens. Upper and lower limits of these populations determined at the 95% confidence level using standard method of logarithmic transformation.

* Arithmetical.

TABLE 3 : *Bacterial contamination of the upper intestinal in 20 Indonesian children with malnutrition and diarrhoea.*

Organism	Number of isolation 10^4 per ml or more	Range (log_{10} organism per ml)
Coagulase-positive Staphylococci	8	0 — 7.8
Other Staphylococci	9	0 — 7.0
Haemophilus influenzae	—	—
Corynebacterium spp.	—	0 — 3.9
Candida spp.	8	0 — 6.3
Escherichia coli	7	0 — 8.0
Klebsiella spp.	3	0 — 8.9
Pseudomonas spp.	5	0 — 8.6
Salmonella parathyphi B	2	0 — 4.4
Shigella spp.	1	3
Other enterobacteria	8	0 — 8.7
Aerobic lactobacilli	4	0 — 5.7
Streptococcus faecalis	—	0 — 3.5
Alpha Haemolytic Streptococci	4	0 — 6.1
Beta Haemolytic Streptococci	—	—
Other Streptococci	9	0 — 8.0
Anaerobes	6	0 — 8.0
Total flora	18	0 — 9.1

TABLE 4: *Oro-pharyngeal Microflora in Well-nourished and Malnourished children. (Results indicate the percentage rate of isolation of organisms in the categories)*

Normal oro-pharyngeal micro-organisms											
	Staph.* pyogenes	Staph.* saprophyticus	Micrococcus	Alpha haemolytic Streptococcus	beta haemolytic Staph.	Streptococcus other	Haemophilus spp.*	Bacillus	Neisseria spp.	Diphtheroids	Candida spp.*
Caucasian (42)	36	50	2	67	12	20	9	12	38	9	7
Australian aborigines (28)	25	79	7	57	7	14	4	4	43	21	14
Indonesians (28)	43	57	4	75	—	11	14	4	4	—	36

* may be pathogenic when present in large numbers.

TABLE 5 : *Gastric and small-intestinal colony counts per ml of Candida spp. in patients with and without antibiotic treatment*

	Treated		Untreated	P
Caucasians	Gastric	2.1 (8)	1.4 (19)	$0.2 < p < 0.25$
	Intestinal	0.8 (8)	0.8 (19)	$0.3 < p < 0.35$
Australian aborigines	Gastric	2.6 (10)	3.3 (20)	$0.2 < p < 0.25$
	Intestinal	2.3 (10)	2.9 (20)	$0.2 < p < 0.25$
Indonesians	Gastric	3.9 (19)	2.6 (8)	$0.25 < p < 0.3$
	Intestinal	2.1 (19)	1.6 (7)	$0.3 < p < 0.35$

Results indicate populations of Candida spp. expressed as the log of the mean viable colony counts per ml of specimens. Figures in parentheses indicate the number of studies.

TABLE 6 : *Gastric and small-intestinal colony counts of Candida spp. per ml expressed as the log_{10} of mean viable colony count*

		Gastric		Small - Intestinal	
		Mean Colony Count	P	Mean Colony Count	P
Caucasians	(27)	0.8	—	1.6	—
Australian aborigines	(30)	2.8	< 0.0005	3.1	< 0.025
Indonesians	(27)	2.0	< 0.025	3.5*	< 0.01

* 26 studies.

TABLE 7 : *Species of Candida isolated from small intestinal aspirates in numbers greater than 10^3/ml*

Subject		C. albicans	C. tropicalis	C. parapsilosis	C. krusei	C. guillier Mondii
Caucasians	(27)	2	1	2	7	—
Australian aborigines*	(30)	6	7	2	1	1
Indonesian	(27)	6	5	17	—	—

TABLE 8 : *Percentage of isolation of Candida spp. and age of the patients*

Age (month)	Total cases	Candida	Percentage (%)
0 — 1	538	177	33
1 — 6	216	70	32.4
6 — 12	131	56	42
12 — 24	74	43	58
24 — 36	42	24	57
Total	1001	370	37

TABLE 9 : *Percentage of isolation of Candida spp. and nutritional state of the patients*

Nutritional State	Total cases	Candida (+)	Percentage (%)
Normal	415	146	34.9
Underweight	482	180	37.4
P. E. M.	104	44	42.1

TABLE 10 : *Percentage of isolation of Candida spp. and the administration of antibiotics*

	Number of cases	Candida	Percentage (%)
Without antibiotic	660	224	34
With antibiotic	341	146	43

TABLE 11 : *Effect of Nystatin on intestinal candidiasis*

Number of cases	Duration of treatment (days)	Result of treatment		Cure rate
		Candida neg.	Candida pos.	
370	5	323	49	87.3%
47	7	28	19	94.3%
19	10	19	0	100 %

neg. = negative pos. = positive

TABLE 12 : *Small intestinal microflora in 10 malnourished Indonesian children*

	Gram-positive bacteria	Candida	Enterobacteria	Faecal bacteria	Anaerobes	Total organisms
No. of isolations $> 10^3$/ml	2	3	3	5	9	9
Range of isolations in all patients	0-1.4x10^4	0-5.6x10^4	0-2.1x10^5	0-2.1x10^5	0-9.0x10^6	0-9.2x10^6

TABLE 13 : *Candida-killing capacity (expressed as percentage of Candida killed) according to nutritional status expressed as a percentage of standard weight for age compared with the 50th centile of the Harvard Standards (Lancet, 1970)*

	Well-nourished group	Undernourished group	Malnourished group
Percentage of candida killed	44.5	17.6	13.7

Candida spp. were found in intestinal aspirates from only six of the Caucasian patients. Large numbers of Candida spp. (10^4 to 10^8 per ml) occurred in 23 aboriginal children and 12 of the Indonesian children.

The rate of isolation was significantly increased in each instance in the latter two groups (Table 6).

There were considerable differences in the species of Candida found in the three groups of patients; these are indicated in Table 7. In individual patients multiple isolations were common. In the Caucasians, isolations of Candida albicans were uncommon; the most common species found was C. krusei. Isolations of Candida albicans and C. tropicalis were common in the aborigines and Indonesians; C. parapsilosis was very common in the latter group.

4. Isolation of Candida spp. from the stool of infants and children with diarrhoea (Sunoto, R. Henny Purboyo, Adnan S. Wiharta and R. Sutejo, 1979).

4.1. Percentage of isolation of Candida spp. and age of the patients.

1001 infants and children less than 3 years of age from Out Patient Department, Department of Child Health,

Medical School, University of Indonesia/ Dr. Cipto Mangunkusumo General Hospital, Jakarta with diarrhoea of more than 5 days' duration were studied.

From Table 8 it can be seen that the frequency of Candida in children is increasing with the age i.e. the older the child, the higher the percentage. The figures for the age of less than one month, 1 - 6, 6 - 12, 12 - 24 and over 24 months were 33%, 32.4%, 42%, 58% and 57% respectively.

4.2. Percentage of isolation of Candida spp. and nutritional state of the patients.

The percentage of the isolation of Candida spp. increased clearly with the degree of the nutritional state. The figures for the well-nourished, underweight and PEM were 34.9%, 37.4% and 42.1% respectively.

4.3. Percentage of isolation of Candida spp. and the administration of antibiotics.

In contrast to the previous study (Gracey et al., 1973), in this present study it can be seen that the percentage of the isolation of Candida spp. is higher in antibiotic groups than in non antibiotic groups. The figure is 43% in the antibiotic group compared to 34% in non antibiotic group.

4.4 Effect of nystatin on intestinal Candidiasis.

Treatment with nystatin in patients with positive Candida spp. for 5,7, and 10 days revealed a cure rate of 87.3%, 94.3% and 100% respectively. No resistant case was found to nystatin.

5. The use of a simple duodenal capsule to study the upper intestinal microflora (M. Gracey, Suharyono and Sunoto, 1977).

By using a simple duodenal capsule, i.e. the paediatric enterotest capsule (Enterotest TM-Health Development Corporation/HEDECO, Polo, Alto, California), in 10 malnourished children aged from 18 months to 10 years, with the mean age of 6 years revealed 3 out of 10 harboured profuse fungal mycelia suspected Candida spp. The number of the isolations was more than 10^3/ml with the range of isolation of all patients $0 - 5.6 \times 10^4$ per ml.

6. Defective-Candida-killing in childhood malnutrition (R. Tuck, V. Burke, M. Gracey, A. Malajczuk aand Sunoto, 1979).

A study of the Candida-killing-ability of polymorphs in 67 well-nourished and malnourished patients revealed that malnourished children have an impaired Candidacidal activity of leucocytes, a finding which runs parallel to the higher rates of isolation of Candida spp. from throat secretions.

In well-nourished subjects the mean Candidacidal ability was 44.5%, whereas in the moderately undernourished group it was 17.6% and in the severely malnourished group it was 13.7%.

Discussion

From the results of the aforementioned studies it can be seen that intestinal candidiasis occurred commonly in malnourished as well as in well-nourished children with diarrhoea.

Little information is available about the occurrence of yeasts in the alimentary tract in health and disease.

Drasar et al. (1969), studied 42 healthy adults, found yeasts in gastric contents of only 5 individuals and in upper intestinal contents of only one.

In the present study, Candida spp. are uncommon in the relatively well-nourished Caucasian children since there are only 6 (22%) isolations out of 27 patients.

In a study of diarrhoeal disease of children, Kozinn and Taschdjian (1962) found a frequency of 24%, whereas Ono Dewanoto et al. (1968) and Suprapti Thaib et al. (1968) found a figure of 38.1% and 38.6% respectively, in their study of diarrhoeal disease of children less than 2 years of age.

This finding was approximately similar with our finding in 1001 children less than 3 years of age with diarrhoeal disease of more than 5 days duration with all kinds of nutritional stage, i.e. 37% (Table 8).

In normal neonates, Simon and Klose-Gerlich (1969) in Kiel, Germany, found a frequency of 1 - 2% only, whereas in low birth weights it was 20%. In infants and children who were sick and received antibiotics the figure is higher i.e. 30 - 60%.

In our previous study (Table 5), the difference of the frequency in the antibiotic and non-antibiotic groups is not significant. But this study was done in 21 children only.

In our further study with 1001 children less than 3 years of age, it revealed that the figure in the antibiotic group is much higher than in non-antibiotic group i.e. 43% vs 34%.

Compared to the findings of isolation of Candida spp. in well-nourished children, there was a very significant increase in the number of isolations and mean colony counts of Candida spp. in the oropharyngeal cavity, gastric contents and stools from malnourished children, which could be seen in the tables 2, 4, 6, 7 and 9.

Mata et al. (1972) found also yeasts in the gastric and intestinal contents in the majority of children they studied with malnutrition and diarrhoea; isolation was less common in malnourished children without diarrhoea.

Apart from indicating the frequency of isolation of Candida spp. in malnourished children, the present study documents in some detail the species found.

Interestingly, the recognized pathogens, Candida albicans, were found more commonly in the malnourished children, as it was the case with C. tropicalis and

C.parapsilosis. Little is known of the pathogenicity of these other species, largely because most studies have not attempted to determine their occurrence in various disease states and have designated that C.parapsilosis, at least, has an important pathogenic potential.

The present study indicates that species of Candida occur commonly in the gastrointestinal tract of children with malnutrition, possibly because of impaired immunological surveillance.

In our study of candida-killing-ability in well-nourished and malnourished group (Table 13) using the method of Lehrer and Cline (1969) with the modifications of Xanthou et al. (1975) to minimize the volume of blood taken from the children, it was shown that in malnourished children the candidacidal activity of their leucocytes was significantly reduced. This finding runs parallel to the higher isolation rate of Candida spp. from throat swabs. Organisms isolated from the upper small bowel are similar to the flora of the oral cavity and our previous study suggest that Candida spp. are likely to be found in the upper intestine of these children (Gracey et al., 1973).

Our finding showed that polymorphs from malnourished children have an impaired ability to kill candida and extend the observations that the ability to kill Staphylococcus pyogenes is reduced (Seth and Chandra, 1972). These mechanisms may contribute to the significantly increased incidence of candidal infections in malnourished children. And as has been studied by Burke et al. (1977) and Thelen et al. (1978) this overgrowth of Candida spp. in the small-intestine may contribute to the production of diarrhoea by their effects on intestinal absorption of sugar and water.

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