ORIGINAL ARTICLE

Lipiodol Absorption Test with a Chloroform Solution Indicator

by

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Abstract

The incidence of fat malabsorption in neonates with diarrhea was investigated from July until November 1975, by the lipiodol absorption test which used a chloroform solution indicator.

In our study we got the following results :

— *45.9% out of 24 neonates with diarrhea had fat malabsorption. This broke down to 9.1% with mild, 63.6% with moderate and 27.3% with severe fat malabsorption.*

— *45% of the patients had mild dehydration and 50% had severe dehydration with fat malabsorption.*

Usually there is a correlation between the microscopic examination of stools and the lipiodol absorption test.

The employment of the lipiodol absorption test with a chloroform solution indicator is simpler and easier than with an amylum solution indicator.

Received 15th April 1981.

Introduction

The incidence of gastroenteritis in neonates is very high in Indonesia (Aswitha Budiarso et al, 1977; Purnomo Surjantoro et al, 1978; Immanuel Mustadjab et al, 1979).

A high percentage of diarrhoeal illness in neonates is caused by the high prevalence of infections and the number of high risk infants especially low birth weight infants.

To improve and solve the problem of gastroenteritis in neonates, beside the factors of dehydration and infection, the problem of malabsorption must be given attention.

The problem of fat malabsorption in Indonesia is very commonly found in neonates and in children usually in relation to gastroenteritis, PCM, and low birth weight infants. (Suharjono et al, 1977 and Immanuel Mustadjab et al., 1979).

The purpose of this study is to show the incidence of fat malabsorption in neonates with diarrhea which was examined with the lipiodol absorption test using a chloroform solution as an indicator.

Material and methods

The problem of fat malabsorption has been examined in the neonatal unit, Department of Child Health, Medical School, University of Sam Ratulangi, Gunung Wenang General Hospital, Manado from July until November 1975.

The diagnosis of fat malabsorption was established with a microscpic examination of the stool in a 1% eosin solution and the lipiodol absorption test with a chloroform solution indicator.

The method of the lipiodol absorption test is as follows :

1. For each sample of urine to be tested, 7 tubes were placed in a rack.
2. ½ cc of destilled water was placed in tubes 2 to 7 and ½ cc of the sample was placed in tubes 1 and 2.
3. After mixing, ½ cc was removed from tube 2 and placed in tube 3.

Again, after mixing, ½ cc was removed from tube 2 and placed in tube 4, and so on, until finally, ½ cc was removed from tube 7 and discarded.

This procedure supplied ½ cc of undiluted urine and dilutions of 1 : 1 through 1 : 32.

4. To each tube were added 10 drops of concentrated nitric acid.
5. The tubes were then shaken and allowed to stand for 5 minutes, at room temperature.
6. Then to each tube ½ cc of chloroform solution was added.
7. The reaction was positive if a violet colour developed.

Patients without fat malabsorption gave a positive reaction (violet colour due to the reaction of iodine with the chloroform solution) in dilutions more than ¼ (⅛ — $^1/_{16}$ — $^1/_{32}$).

Patient with fat malabsorption gave no reaction (no violet colour) in all dilutions or a positive reaction (violet colour) only in undiluted or up to ¼ dilutions of urine.

Results

The results are summarized in the following tables :

TABLE 1 : *Incidence of fat malabsorption.*

Sex	Degree of fat malabsorption				Normal
	Mild	Moderate	Severe	Total	
Male	1	3	1	5	8
Female	—	4	2	6	5
Total	1 (9.1%)	7 (63.6%)	3 (27.3%)	11 (45.9%)	13 (54.1%)

TABLE 2 : *Correlation between dehydration and fat malabsorption.*

Degree of dehydration	Degree of fat malabsorption				Normal
	Mild	Moderate	Severe	Total	
Mild	—	—	—	—	—
Moderate	1	6	2	9 (45%)	11
Severe	—	1	1	2 (50%)	2

TABLE 3 : *Correlation between maturity and fat malabsorption.*

Maturity	Degree of fat malabsorption				Normal
	Mild	Moderate	Severe	Total	
• L B W Infants	1	4	2	7 (53.8%)	6
Term	—	3	1	4 (36.3%)	7

• LBW = Low birth weight.

TABLE 4 : *Correlation between the microscopic examinations of stools and fat malabsorption.*

Microscopy of stool	Degree of fat malabsorption				Normal
	Mild	Moderate	Severe	Total	
—	—	—	—	—	11
+	1	—	—	1 (9%)	9
++	—	5	1	6 (54.6%)	—
+++	—	2	2	4 (36.4%)	—

Discussion

Because the method of Van de Kamer et al (1949) was not feasible, we used the lipiodol absorption test as a procedure for diagnosing fat malabsorption in addition to the microscopic investigation.

The use of lipiodol as a test substance, with a measurement of iodine appearing in the urine, has been previously reported by Silverman and Shirben (1955), Delory et al, (1956), and Jones and Sant' Agnese, (1963).

Lipiodol consists of poppy seed oil with hydrochloric acid to produce an oil containing 40 percent iodine. The iodine is firmly attached to the unsaturated double bound of the fat and remains bound until after absorption. During the process of transport and assimilation of the fat, the iodine is split and excreted in the urine. The degree of recovery of iodine in the urine reflects the percentage of absorption of lipiodol which in turn depends on the individuals ability to absorb dietary fat.

Lipiodol absorption, like dietary fat absorption, is impaired by a defect in any of the several factors necessary for fat digestion and absorption including the presence of bile salts, pancreatic lipase and an adequate amount of intact small intestine.

According to the presence of the colour in the urine as a result of the reaction between the iodine and the material of the indicator, we can know the presence of iodine which is excreted in the urine and vice versa.

In our study we used a chloroform solution as an indicator in the lipiodol absorption test.

According to the physical and chemical properties of iodine, the iodine can react with the polar and non-polar solution and give a specific colour in the solution as follows :

- Iodine + amylum solution ——→ blue colour.
- Iodine + polar solution (chloroform, carbon disulfide, carbon tetrachloride) ——→ violet colour.
- Iodine + non-polar solution (ethyl-alcohol, ethyl-ether) ——→ brown colour.

When we compare the method of the lipiodol absorption test with an amylum solution indicator in contrast to the chloroform solution, we find that the chloroform solution is easier and simpler. Therefore, this method can be used in the peripheral hospitals.

It has been well-known that many factors may play a role in causing fat malabsorption :

1. Absence or deficiency of lipase.
2. Absence or deficiency of conjugated bile salts.
3. Disturbance in the formation of chylomicron due to the abnormality of the mucosa of the gut.
4. Abnormality of lymphatic vessels (Suharjono et al, 1977).

The incidence of fat malabsorption in Indonesian infants is probably due to various important factors, i.e. :

1. The high rate of low birth weight infants.
2. The high incidence of infections.
3. The high incidence of undernourished infants.

In our examination we arrived at the following results :

- 45.9% out of 24 neonates with diarrhea had fat malabsorption. This broke down to 9.1% with mild, 63.6% with moderate and 27.3% with severe fat malabsorption (Table 1).
- 45% of the patients had mild dehydration and 50% had severe dehydration with fat malabsorption (Table 2).
- 53% of low birth weight infants and 36.3% of term infants had fat malabsorpton (Table 3).

Aswitha Budiarso et al., (1977) found the incidence of fat malabsorption in neonates with diarrhoea as follows; 89.5% out of 76 low birth weight infants and 60.9% out of 121 term infants.

The high incidence of fat malabsorption in low birth weight infants is due to many factors.

The capacity of the gut to absorb fat is limited in neonates and usually in low birth weight infants.

In low birth weight infants all organs are still immature and their function is not yet optimal.

Due to the immaturity of the pancreas and liver, theoretically a lipase and saturated bile salts deficiency may occur and the chylomicron formation may be disturbed too.

Infection of the gut especially by EPEC is very common in neonates, and this infection can cause diarrhoea due to damage of the mucosa, resulting in atrophy

of the villi. The subsequent disturbance of chylomicron formation results in fat malabsorption.

Usually there is a correlation between the microscopic examination of stool and the lipiodol absorption test. The microscopic examination of stool, when compared to the lipiodol absorption test shows an error of only 15 percent. On microscopic examination of the stools we found an error of 9%; one out of 11 patients had a 1 + result and the rest were 2 + (54.6%) and 3 + (36.4%) (Table 4). This suggests that notwithstanding the possiblity of error, the microscopic method remains a valuable screening technique.

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