

Antifertility effect of *Jatropha Curcas L.* seed in guinea pigs

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Abstract: A crude seed extract of *Jatropha curcas* was studied for its claimed anti-fertility effect. The extract was orally administered to matured female albino guinea pigs. It was found to reduce the number of births. This was further confirmed by the anti-implantation and abortifacient effects of the extract observed in this study. The extract was also found to prolong the oestrus cycle of guinea pigs. The diestrus phase was significantly increased, while the oestrus phase was shortened. The weight of the uterus was shown to reduce in animals treated with the extract, while that of the ovaries did not show a significant change from that of the control. These observations suggest that the seeds of the plant possess antifertility activity. [Ethiop. J. Health Dev. 1997;11(2):145-148]

Introduction

Family planning has received increased attention world-wide because of the ever-expanding population that will lead to economic and health impact on the family and society as a whole (1,2). One of the means through which developed countries can control their population size is by making available modern oral contraceptives to the population. This, however, may not be affordable to the majority of the population in developing countries. Hence, the need arose for exploration of the natural products which have been used traditionally for years.

In search for anti-fertility agents, some have been screened with demonstrable positive results in either of the sexes. For example, the seeds of *Carica papaya* (3) and the roots of *Moringa oleifera* (4) were found to be effective anti-fertility agents in male and female rats, respectively. Besides the screened antifertility plants in laboratories, there are many others traditionally claimed to have this effect as well. Among the many traditionally claimed antifertility plants in Ethiopia is *J. curcas* (commonly called physic nut or pig nut and, in Amharic yefernj gulo). It is a small tree belonging to the family Euphorbiaceae, and found distributed throughout the tropics, including in West, East and southern Africa (5). It thrives best in areas with a rainfall not less than 1000 mm/year and temperatures not exceeding 40°C (5). The fruiting period of the plant is between July and August (5). Its various parts are traditionally employed for different ailments. For instance, the root is employed in infantile tetanus, dropsy, sciatica, paralysis, and skin diseases (6); its leaf is used in jaundice, as a mouth wash, in convulsion and as an anti-helminthic agent (5); and the seed is used as purgative (6,7), as abortifacient (8), and as contraceptive (7).

The constituents of the plant have been studied and were found to be fixed oil consisting of mainly glycerides of stearic, palmitic, myristic, oleanic and curcanoleic acids (5). The juice contains tannin (8), and the seed contains toxalbumin (8).

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The aim of the present study is to investigate the antifertility effect of its seed in female albino guinea pigs.

Methods

Extraction of the plant material: The dried seeds of *J. curcas* were collected from a town of Shewa Robbit, North Shewa, in the month of September. These were further dried in the shade and ground to powder. The powdered material was macerated with water for 24 hours and filtered with Wattman filter Paper (No 1). The filtrate was lyophilized, weighed and then reconstituted to obtain different concentrations for use in the study.

Animal preparation: Six female albino guinea pigs per group were employed. The guinea pigs were 3 - 5 months old and weighed 500 - 600g each. All the animals were made to acclimatize for about two weeks prior to the commencement of the experiments.

Preliminary screening for the antifertility effect: Two groups of matured female guinea pigs were selected for this experiment. One of the groups served as a control and received the vehicle by intra-gastric intubation. The other group received 0.3g/kg/day of the crude extract by the same route of administration starting a week earlier before mating. Both groups were then allowed to mate with matured male guinea pigs (one male for two females) according to the method of Bertholet (9). The number of births was then determined after the completion of one gestation period in both groups as described by Anokbonggo (10).

Anti-implantation and Abortifacient Effects: Two groups of the matured female guinea pigs were employed for this experiment. One of the groups received 0.3g/kg/day of the extract by intra-gastric intubation for ten days before mating, and was continued after mating for twenty seven days. On the 33rd day after mating, the animals were sacrificed. The control group received the vehicle by the same route, and were sacrificed on the same day. The number of implantation sites and that of death vis-a-vis alive fetuses were recorded for both groups according to the methods described by Dhwan and Srimal (11), and by Briggs and Diczfalusy (12).

Effect on oestrus cycle: Six matured female guinea pigs were employed. Vaginal smear was taken from each animal, and examined under a microscope every day for about five cycles (a total of 27 days). The length of oestrus cycle and those of the different phases were determined in accordance with the method described by Briggs and Diczfalusy (12). The seed extract (0.3g/kg/day) was then administered to all guinea pigs every day for same period, i.e., 27 days, and the same parameters were determined.

Effect on the weight of ovaries and uterus: Two groups (test and control) of matured female guinea pigs were employed. To the test group, the crude extract of the plant material (0.3g/kg/day) was administered by intra-gastric intubation whereas, the control was given the vehicle by the same route. After 27 days, all the animals were sacrificed and the weights of the uterus and of the ovaries were then taken according to the method described by Briggs and Diczfalusy(12).

Statistical analysis: The results were analyzed statistically, and student t test was employed to test for level of significance. All data were expressed as Mean \pm Standard Error of the Mean (SEM).

Results

The crude extract of *J. curcas* seed was found to reduce significantly ($p < 0.05$) the number of births given by a guinea pig varied from 2.83 ± 0.75 to 0.50 ± 0.86 . The extract was also observed to reduce significantly ($p < 0.05$) the number of sites of implantation (Table 1).

Table 1: **Effect of *Jatropha curcas* seed (0.3g/kg/day for 27 days) on the number of sites of pregnancy, and that of dead and alive fetuses.**

	Control	Extract
No. of sites of pregnancy	3.20 ± 0.84	* 2.10 ± 0.52
No. of dead fetus	0.20 ± 0.45	$1.60 \pm 0.50^{**}$
No. of alive fetus	3.00 ± 0.80	$0.60 \pm 0.49^{**}$

n = 6, Mean \pm SEM, * $p < 0.05$, ** $p < 0.01$.

The number of dead fetuses in the experimental animals was significantly higher than those that are alive, while the reverse was true for the control animals as indicated in Table 1 ($p < 0.01$). Table 2 shows that the extract prolonged significantly ($p < 0.01$) the oestrus cycle. The same table shows a significant increase in the diestrus phase and decrease in the oestrus phase ($p < 0.01$) by the seed extract. The weight of the uterus was significantly reduced ($p < 0.01$) by the extract, while that of the ovaries did not show a significant change from that of the control (Table 3).

Table 2: **Effect of *J. curcas* seed (0.3g/kg/day for 27 days) on the oestrus cycle.**

	Control	Extract
Oestrus cycle (day)	5.75 ± 0.01	14.25 ± 1.19**
Proestrus phase(day)	0.92 ± 0.26	0.13 ± 0.18
Oestrus phase (day)	2.17 ± 0.26	0.27 ± 0.25**
Metooestrus phase(day)	0.58 ± 0.20	0.60 ± 0.42
Diestrus phase(day)	2.08 ± 0.26	13.00 ± 1.17**

n = 6, Mean ± SEM, ** $p < 0.01$.

Discussion

The reduction in the number of births with the crude extract of *J. curcas* seed suggests its possible antifertility activity.

The decrease in the number of births may be attributed partly to the anti-implantation effect of the extract as demonstrated by the decrease in the implantation sites in this study. This could also be attributed to the abortifacient effect of the extract as demonstrated by the increased number of dead fetuses in animals treated with the extract.

Table 3: **Effect of *Jatropha curcas* seed (0.3g/kg/day for 27 days) on the weights of the uterus and ovaries in nonpregnant guinea pigs.**

	Control	Extract
Uterus wt. (g)	1.062 ± 0.014	0.728 ± 0.129**
Ovaries wt.(g)	0.242 ± 0.087	0.251 ± 0.058

n = 6, Mean ± SEM, ** $p < 0.01$.

The prolonged oestrus cycle with an increase in the diestrus phase (safe period) and a decrease in the oestrus phase (unsafe period) in animals treated with the extract may explain the remote chance of becoming pregnant. This further supports the suggestion for the antifertility effect of the extract.

The reduced weight of the uterus in non-pregnant animals may explain for the possible role of the extract on hormonal changes taking place in the animal. This may give a clue to the possible mechanism (s) of action of the extract.

As the present study concentrates only on the efficacy of the aqueous extract of the seed of *J. curcas* for its anti-fertility effect, further studies should be undertaken on the mechanism(s) of action, toxicity tests and Pharmacokinetics.

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Original article

Indigenous weaning foods: Hygiene and diarrhoeal diseases in rural Ethiopian setting, Jimma Zone

Abstract: A community based Microbiological study of Weaning Foods was conducted from Nov. 1994 to August 1995 in six Peasant Associations, in Jimma Zone, South West Ethiopia. The results of the study indicate that infants and children in the study community are ingesting highly contaminated foods. Fifty five percent of the weaning foods harbored bacterial counts over 2×10^6 /ml of samples, and coliform bacteria were isolated from over 50% of these foods. The invariably high contamination of weaning foods was significantly associated with unsafe water supply, storage at ambient temperature for long time, unsatisfactory methods of cleaning feeding utensils. Poor domestic and personal hygiene observed in the households surveyed could provide ample opportunity for communication of fecal organisms to food and water either directly or indirectly. There was no statistically significant association between bacterial contamination of weaning foods and diarrhoeal episodes in children ($P > 0.05$). Educating the rural community, and improving over all hygienic conditions in the home environment is crucial for improving food hygiene and reduce the risk of diarrhoea from contaminated foods. [*Ethiop. J. Health Dev.* 1997;11(2):149-156]

Introduction

There is a high incidence of childhood diarrhoea in many developing countries despite intensive activities of diarrhoeal disease control programmes (1). Numerous studies have been conducted to examine the relationship between feeding practice during the weaning period and the risk of diarrhoea and malnutrition (2). Infants are at greatest risk of diarrhoea when foods other than breast milk are first given (3). Since the standard of personal hygiene and public sanitation are low in many developing countries, the increased risk of diarrhoea observed with the introduction of weaning foods suggests contamination of these foods with pathogenic microorganisms (2-4). Most diarrhoea episodes have been associated with organisms such as *Campylobacter jejuni*, enterotoxigenic and enteropathogenic *Escherichia coli*, Shigella, Salmonella, rotavirus, and protozoa parasites (5,10). These organisms appeared to be transmitted to infants in the home through feces contaminated water and food and direct person to person contact (5). Because of the difficulty of isolating enteric pathogens from foods most studies have either used the enumeration of "indicator" organisms as evidence of the potential for transmission of enteric pathogens or aerobic bacterial count to indicate level of contamination (2,11).

Breast feeding is the safest practice since breast milk is free of contamination and its protective value against diarrhoeal morbidity and mortality is well recognized (2). In most traditional communities in the developing world breast feeding is practiced almost universally. However, after an age of 4-6 months, breast milk alone is not adequate to satisfy the nutrient requirements recommended by the Food and Agricultural Organization and the World Health

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Organization (6). This means that gradual introduction of foods other than breast milk is needed for infants to grow well. Safe preparation and storage of food should be a priority during weaning (7). The production of a sterile weaning food for a baby depends on a number of factors which include the effectiveness of utensil sterilization, the degree of contamination of the original food material, the adequacy of measures taken to sterilize the weaning foods and to keep them sterile until they are given to the baby, and improved personal and domestic hygiene in the home environment (8). Studies in Africa and other countries have documented that indigenous weaning foods including animal milk are grossly contaminated (8-11).

A recent study in Addis Ababa, Ethiopia (12) has examined infant feeds and feeding utensils and recovered pathogenic microorganisms from these feeds and feeding utensils. The above study considered subjects attending hospitals and clinics in urban setting and samples of infant foods were taken in the hospital and clinic environments. Our study is a community based survey in the rural

Ethiopian setting. It is a microbiological survey which included a house to house visit to observe hygienic conditions of the home environment and the weaning foods. The specific objectives of the study are: 1. to investigate methods of preparation and storage of indigenous

weaning foods and determine the possible point and source of contamination, 2. to determine the level of bacterial contamination of weaning foods, 3. to observe hygienic condition of home environment in connection with contamination of weaning foods, 4. to determine the association of contaminations of weaning foods to the incidence of diarrhoeal diseases in the study population.

Methods

1. *Area of study:* Six Peasant Associations in three districts; Yebu, Serbo, and Seka-Chokorsa (20-25 km away from Jimma town) in Jimma zone, South West Ethiopia, were selected for the study. The criteria for selecting these peasant associations in these districts were:

1.1 Their proximity to the microbiology laboratory of Jimma Institute of Health Sciences (JIHS).

1.2 A positive attitude of the Community to such a study: These are sites for Community Based Training Program (CBTP) of JIHS.

1.3 Organization of the villages by Villagization Programme.

1.4 Availability of transport to the area.

2. *Subjects:* Subjects were all households in six peasant associations with mothers of infants and children between 2 and 24 months of age and who give their babies supplements in addition to breast milk.

3. *Interviewing of mothers:* Mothers or care-takers (family members who is/are responsible for the child in the absence of the mother) of children 2 to 24 months were interviewed at their homes with a questionnaire used in a similar study (12). The questions included level of mothers' education, method of feed preparation, methods of cleaning feeding utensils, duration feeds are kept from preparation until consumption, etc. The language used to interview the mothers was their vernacular (Oromigna). The interviewer was a senior Laboratory technician who speaks the language. A mother/care taker was first explained about the purpose of the study and interviewed if she agreed to provide the necessary information and samples required for the study. In addition to interviewing, the research group (Lab. technician, either of the authors) observed the sanitary condition of the home environment such as waste disposal, personal and domestic hygiene (eg. hand washing during handling weaning foods etc.). The study did not include exclusive breast-fed children.

4a. *Collection of Food samples:* Depending on the available weaning food types, solid, semisolid or liquid foods ready for consumption were sampled by following standard bacteriological techniques (13). Samples were collected in the mornings from 9:00 a.m. to 11:00 a.m. Most liquid and semi-solid foods were collected within 2 to 3 hours of preparation. Solid foods usually prepared for consumption by adults were also given to children over 1 year of age (in which case time elapsed between preparation and sample collection may range from 1 to 48 hours). Raw food items were also collected from some households. All samples were then placed in sterile screwcapped bottles and were kept in cold chain system until processed in the laboratory.

4b. *Rectal Swabs:* Rectal swabs were collected by a laboratory technician by inserting sterile cotton swab into the rectum of infants and children whose weaning food samples were taken. The rectal swabs were placed in Cary-Blair (Difco) transport media and kept in the cold chain system.

5. *Processing of the specimens:* The samples were processed within 2-3 hours of collection in the Microbiology Laboratory of JIHS. Food samples were processed for (i) isolation and identification of possible pathogens and (ii) for total aerobic colony counts using bacteriological techniques previously described by Banwart (14). The rectal swabs were cultured on Primary isolation media MacConkey and SSA and growth was examined and tested by biochemical methods (15). Solid or semi-solid foods were blended with sterile saline as described by previous workers (10).

6. *Enumeration and Identification:* Bacterial colonies on standard plate count agar plates were counted using automatic colony counter and recorded as colony/ml of samples. Growth of enteric pathogens were examined on MacConkey agar and standard biochemical tests previously described

by Cheesbrough (15) were used in the primary identification of the growth in the appropriate media. Specific antisera were used as confirmatory test in identification of specific pathogens such as enteropathogenic *E-coli* and *Shigella*.

7. *Data Analysis:* Analysis of data generated by microbiological procedures and information obtained by questionnaire were processed using EPI-INFO (version 5) statistical software. Chi square test was used to determine associations between different variables.

Results

Five hundred sixty five households were included in the study conducted from Nov. 1994 to August 1995. Out of these, 562 (99.5%) households could provide the necessary information and samples required for bacteriological analysis, the remaining 3 (0.5%) households could not provide complete information and were excluded from the study. The majority, 481 (85.6%) of the households in the study area live in thatched houses and the remaining, 81 (14.4%) dwell in corrugated iron sheet houses. Of the 562 households, 477 (84.9%) live in the same room with cattle, 63 (11.2%) of the households keep their cattle in separate rooms, and the rest do not possess any cattle. Four hundred and thirteen (over 73%) of the households do not possess pets, and only 149 (26.5%) possess dogs and cats.

Table 1: **Educational level of mothers in the studied community Jimma Zone, Nov. 1994 - Aug. 1995.**

S.No.	Level of Education Mothers	Total Number of	Percent of Total
1	Illiterate	118	118
2	Literate*	376	376
3	Grade 1-6	54	54
4	" 7-9	9	9
5	" 9-12	5	5
Total		562	562

* able to read and write through informal education

The majority, 441 (78.5%) of the households get water for domestic purposes from unprotected springs, rivers, wells, and ponds, and only 77 (21.5%) obtain water from protected springs. Sixty seven percent of the mothers have been exposed only to literacy level of education and less than 1 percent have secondary education (Table 1). From among cereals, Teff (*Eragrostis teff*) used by 145 (26%), maize by 107 (19%), sorghum by 34 (6%) of the households to prepare weaning foods. Fresh cow's milk is used by 128 (22.8%), and commercial flour, Dube and Fafa by 77 (13.7%) and 7 (1.2%) of the households respectively (Table 2). Breast feeding is universal (99.6% were breast-fed) in the study population and other foods are given to supplement breast milk beginning 2 months

Table 2: **Types of cereals from which weaning food are prepared versus bacterial counts /ml of samples, Jimma zone, Nov. 1994 - Aug. 1995**

Ser. no	Food Item	Colony counts					
		10 ³	10 ⁴	10 ⁵	10 ⁶	10 ⁷	Total
1	Sorghum	3	3	8	12	8	34
2	Maize	22	3	48	21	3	107
3	Wheat	0	0	2	0	1	3
4	Barley	3	0	3	2	2	10
5	Fresh Cow's milk	6	4	10	30	8	128
6	Teff (<i>E.teff</i>)	16	19	55	34	21	145
7	Mixed cereals	1	0	2	5	5	13
8	Kocho (<i>E. ventricosum</i>)	0	0	6	2	9	17
9	Dube (commercial flour)	3	3	25	18	28	77

10	Fafa (commercial flour)	2	1	2	0	2	7
11	Others (plant juice, roots and tubers)	1	2	4	3	11	21
	Total	57	35	165	127	178	562

of age. Sixteen (2.8%) of the infants in the study households were given supplementary foods such as fresh cow's milk, tea, "abish", at the age of two months, and 59 (10.5%) of the babies are given supplementary foods between the age 3 to 4 months. Fig.1 shows the percent of weaning food types versus aerobic bacterial counts/ml of samples. Of

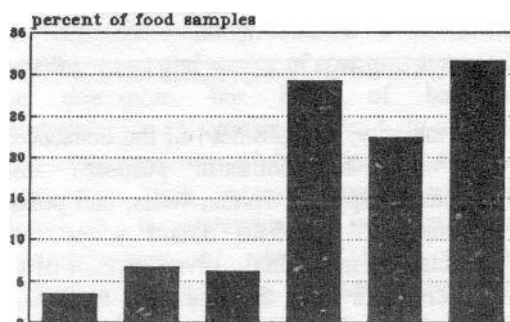


Figure 1: Percent of weaning food samples versus colony count/ml

the 562 weaning food samples 178 (32%) contained bacterial counts 10^7 /ml or more. Table 3 shows methods of cleaning feeding utensils versus bacterial counts in feeds. Methods of weaning food preparation are given on Table 4. Thin/liquid porridge is prepared from different cereals by 138 (24.6%), Cow's milk boiled 65 (50%) of the 128 hosholds who supplement breast milk with cow's milk. Over

Table 3: Methods of cleaning feeding utensils vs. bacterial colony count/ml of food samples, Jimma Zone, Nov. 1994 - Aug.1995.

S. No.	Cleaning Methods	No Growth	$<10^3$	10^3	10^4	10^5	10^6	10^7
1	Boiling utensil (n = 7)	-	-	-	-	2	2	3
2	Wash with boiled water (n = 160)	3	-	4	4	26	36	87
3	Wash with un boiled water (n = 389)	14	1	34	30	136	88	86
4	Wash with soap and unboiled water (n = 6)	1	-	-	1	1	1	2

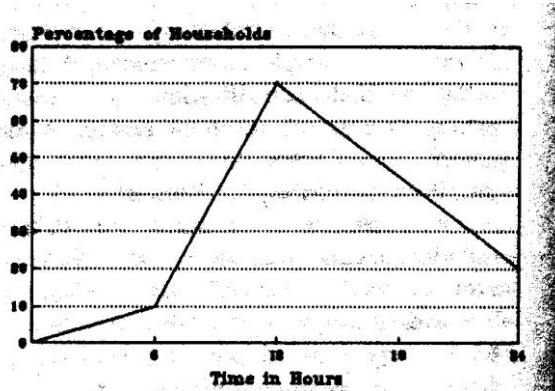


Figure 2: Percent of households VS duration of food keeping before consumption

45% of the mothers give solid food (prepared for adults) to their babies. It was observed that babies over 12 months are given such solid foods. Weaning foods are kept at ambient temperature for 12 hours by 368 (65.5%) of the households only 10 (1.8%) prepare weaning foods for immediate consumption (Fig 2 and Table 5). Over 50% of the babies in the study group are handfed and 117 (20.8%) are bottle fed (Table 6). Most mothers observed in the study do not wash hands, but this observation has not been quantified. Table 7 shows bacterial pathogens isolated from infants and children in the study group. Non enteropathogenic *E.coli* were the most frequent isolate both from diarrhoeal and non diarrhoeal children. No Salmonella or Shigella or enteropathogenic *E.coli* was isolated from 139 (55%) of diarrhoeal and 184 (59%) non diarrhoeal cases.

Table 4: **Methods of Weaning Food Preparation in the studied house-holds, Jimma Zone Nov. 1994- Aug.1995**

S.No	Methods of Preparation	Total No. of house-holds	Percent
1	Thin Porridge	138	24.6
2	Fresh cow's milk served raw	76	13.5
3	Boiled cow's milk	65	11.6
4	Baking as bread or "Enjera"	256	45.6
5	Unprocessed for feeding* (Raw fod items)	25	4.4
6	Others**	2	0.4

N.B * = Any cereal flour collected for analysis ** = Plant or fruit juice given to babies

Discussion

Weaning age children are very prone to infection, therefore safe preparation and storage of weaning foods is crucial to good health of infants and children (16). The results of the present study indicate that babies in this community are commonly ingesting highly contaminated foods. All types of weaning foods are grossly contaminated; fifty five percent of all types of weaning foods harbored bacterial counts over 2×10^6 /ml and coliform bacteria were isolated from over 50 percent of these foods. The present study attempted to determine the possible sources and points of contamination of indigenous weaning foods in the study community. Bacteriological analysis of raw food items from which weaning foods are prepared has shown gross contamination, 60 percent of cereal flour, and 84 percent of animal milk contained aerobic bacterial counts over

2×10^6 /ml of samples. Some of the food samples studied harbored more than one type of bacterial

species. The difference between bacterial counts of raw food items and that of cooked foods is not evident. This may be due to inadequate cooking or unhygienic handling of the cooked feeds. Slightly lower bacterial counts and fewer bacterial isolates were documented in fermented cereal products than in unfermented foods. Recent works on weaning foods indicate that preparations of weaning foods from fermented products reduces risk of contamination (16), but the present study has not focused attention in this area. So the importance of this particular aspect of food preparation in this community

Table 5: **Duration of weaning foods left at ambient temperature until consumption versus bacterial counts in foods, Jimma Zone, Nov.1994- Aug. 1995**

Colony count/ml of weaning foods							
Ser. No.	Duration (hrs)	10 ³	10 ⁴	10 ⁵	10 ⁶	10 ⁷	Total
1	<6	1	2	3	3	1	10
2	6	5	0	4	16	19	54
3	12	38	9	93	85	43	368
4	>12	13	24	55	23	15	130
	Total	57	35	165	127	178	562

awaits future study. The invariably high contamination of weaning foods in this community may be attributed to various factors operating simultaneously. Scarcity and unsafe water supply, poor handling of foods, storage of foods in ambient temperature for long time and poor domestic and personal hygiene may be among the most important contributing factors. Black et al (5) from Huscar, Peru, have found out that milk and foods prepared from cereals were often contaminated. In that study the frequency of contamination was related to the amount of time since initial preparation. In the present study over 60 percent of the households keep weaning foods for 12 hours and above at ambient temperature. There is a significant association between the length of time foods are kept at ambient temperature and the bacterial counts in weaning foods ($P < 0.001$). The present finding agrees with many of the previous works (5,10). Black (10) studied contamination of weaning foods in Bangladesh and has reported that it is a common practice to cook weaning foods in the early morning and storing them at ambient temperature for consumption later in the day. In that study they have observed that such practices encouraged the growth of *E. Coli*. Another study (17) from Jamaica did not observe any correlation of bacteria in the feed samples with the length of time from preparation of the feed by the mother to the time of sampling. Ideally infants should be given freshly prepared food at every meal because storing food at room temperature causes bacteria in the food to multiply. Method of cleaning feeding utensils in the present study shows a statistically significant association with bacterial counts in weaning foods ($P < 0.001$), a similar finding was recorded by Elegbe (9) in Ile-Ife, Nigeria. Previous studies from Addis Ababa, Ethiopia (12), Jamaica (17) and Uganda (8) did not find any significant association between cleaning methods of feeding utensils and bacterial contamination of feeds.

Table 6: **Methods of Feeding children practiced by households, Jimma Zone, Nov.1994- Aug.1995**

S.No	Methods	Total No.	Percent
1	Bottle Feeding	117	20.8
2	Spoon Feeding	10	1.8
3	Hand-Feeding	285	50.7
4	Clay or Plastic Utensil Feeding	150	26.7

Water for preparing weaning foods and for other domestic uses in the majority of the study community is obtained from un protected sources such as streams, rivers, and ponds. Water could be further contaminated at the time of delivery or in the storage container in the households.

Contamination of water and excreta disposal is generally considered to be an important factor in diarrhoeal transmission (18). In the present study source of water and bacterial counts in weaning foods are significantly associated ($P < 0.01$). This is to be expected because unboiled water is used both for preparation of weaning foods and washing of utensils. Hence bacteria may be introduced from contaminated water to weaning foods. Kanem et al (1) have found out that the sources of domestic water are among the main factors which showed significant association with occurrence of diarrhoea.

In the present study it was found that 85% of the households keep cattle in a single room together with people. The rooms were often spoiled with the animal's feces and fecal material of children. Infants were seen crawling freely in the room and compound yard littered with animal and sometimes with human feces. These conditions provide ample opportunity for the communication of diarrhoea pathogens to food and water either directly or indirectly (1,5). So, it appears very difficult to prepare and maintain weaning foods free of contamination in household settings with such poor domestic and personal hygiene. Although most episodes of diarrhoea are caused by people swallowing fecal organisms (18), there is no statistically significant association between bacterial contamination in terms of colony counts in weaning foods, and diarrhoeal episode in the study population ($P > 0.05$). Thirty eight percent of the babies either had suffered an episode of diarrhoea within the last ten days or were suffering during the time of the survey. The remaining majority (62%) had apparently escaped these symptoms inspite of ingesting grossly contaminated foods. In the present study due to limitation of laboratory facilities it was not possible to determine the rate of isolation of enterotoxigenic *E. Coli* and other enteric pathogens such as *Campylobacter jejuni* and rota virus. Enteropathogenic *E. Coli* was isolated almost in the same rate from diarrhoeal and non-diarrhoeal babies. Hibert et al (17) have documented a similar finding in Jamaica. Whether an infant suffers gastroenteritis or not depends not only on the fecal contamination of the weaning food, but also upon the child's ability to resist the establishment of the pathogen in his bowel. When the child has been breast-fed and healthy

Table 7: Enteric Pathogens isolated from rectal swabs of Infants and children in the studied population, Jimma Zone, Nov.1994- Aug.1995

S.No.	Enteric Pathogen	Diarrhoeal Cases (n = 251)	Non-Diarrhoeal Cases (n=311)
1.	Enteropathogenic E-coli	6 (2%)	8 (2.6%)
2.	Non entero-pathogenic E.coli	106 (42%)	118 (38%)
3.	Shigella sp.	-	1
4.	No Pathogen *	139 (55%)	184 (59%)

* No Salmonella, Shigella or enteropathogenic *E.coli* is isolated.

with an intact immune system, gastric acidity, intestinal motility and a normal resident flora, the contaminated food can be ingested with relative impunity. If on the other hand these factors are disturbed then the contaminating organisms can establish themselves and induce diarrhoea (17,19).

There are certain limitations in the present study:

1. samples were collected at different seasons of the year, this may affect the result.
2. the specific time between preparation of weaning food and sample collection could not be indicated by mothers/care takers.
3. although it was observed that most mothers/care takers do not wash their hands before feeding infants, this was not recorded in strict sense and hence was not quantified.

Despite these limitations the present study indicates the hygienic conditions of weaning foods as determined by the level of bacterial counts. The study also gives some clues on possible points and source of contamination of indigenous weaning foods. These includes:

1. contaminated water used for preparation of weaning foods and for cleaning feeding utensils,
2. poor sanitation of home environment as reflected by keeping animals with humans and floors littered with animal dung and human feces,

3. keeping weaning foods for long time at ambient temperature.

Whether the child ingesting such contaminated food suffers from diarrhoea at the time of the study or not, there is a potential risk of contracting diarrhoeal disease at a point in time. Moreover, some of the mothers in the study community introduce supplements earlier than the desirable age (supplements are given in some of the study households at the age of 2 months). Therefore, to effectively reduce transmission of enteric pathogens through weaning foods and prevent diarrhoeal diseases in infants and young children we recommend the following:

1. find out traditional practices that improve food hygiene and work with communities to identify ways of reducing the risk of diarrhoea from contaminated foods,
2. educate the rural community about the links between contamination of weaning foods and diarrhoeal diseases,
3. explain the ill effects of early weaning and emphasize the importance of exclusive breast feeding through continuous health education.

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