

Observation of blood microfilariae during human trypanosomiasis survey in Gambella, south west Ethiopia

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Abstract: While conducting a survey on sleeping sickness during 1989-92 in Gambella, South western Ethiopia, microfilariae of *Mansonella perstans* were detected in the blood of the indigenous population, the Anuaks (1%), and the refugees from Southern Sudan (4.1%). No blood microfilarial infection was detected in resettlers, who arrived in the area from drought affected regions of the country during the 1985/86 resettlement program. Among the diagnostic methods applied, nearly twice more microfilaremic cases (4.1%) were detected by the Miniature Anion Exchange Centrifugation Technique (M-AECT) while only (1.9%) were detectable by Microhaematocrit Buffy Coat Technique (MHBCT) among the refugees. Using the conventional blood film methods (thin and thick smears) only fewer positive cases (1.0%) were detected compared to the above two techniques. Besides a known standard diagnostic methods for blood filariasis, however, the MHBCT seems preferable as field diagnostic technique. Because it is more rapid, simple to operate and does not necessitate as much advanced preparation and sterile condition as M-AECT, and could be a potential diagnostic tool for blood microfilariae. There is a significant difference ($P<0.01$) in age groups 15-30 years among Anuaks and refugees. There is no significant difference ($P>0.01$) in other age groups and sexes among Anuaks and refugees. However, there is a significant difference ($P<0.01$) in over all positivity among Anuaks and refugees. [Ethiop. J. Health Dev. 1997;11(1):1-5]

Introduction

Human trypanosomiasis is known to be endemic in Gambella, South Western Ethiopia (1,2). Meanwhile, different kinds of infectious agents that co-existed with sleeping sickness were also recorded. Among these lymphatic filariasis due to *Wuchereria bancrofti* (3) and a case of *Burgia* species (4) were reported. The area is also known to be endemic for Malaria, Onchocerciasis, Leishmaniasis and *Loa loa*. Some of the vectors of these parasites have also been incremented and described by previous investigators (5,6). The main objective of this study was to evaluate and apply a more sensitive, rapid, simple and reliable diagnostic technique for human trypanosomiasis in the field, therefore scoring for blood microfilariae along with trypanosomes in every Miniature Anion Exchange Centrifugation technique (M-AECT), Microhaematocrit Buffy Coat Centrifugation technique (MHBCT) and the classical methods such as stained blood films (thin and thick smears) and wet film mounting were conducted.

Methods

Study area and population

Gambella is located 800 km from Addis Ababa at latitude 8°N and longitude 34 °E at an altitude of 500 metres. The Baro, Gilo, Aluwero, Akobo rivers and several other small to moderately sized

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rivers criss cross the Region. The mean annual rainfall is 980mm and mean relative humidity is between 45% and 75% (7).

The indigenous population included in this study belongs to the Anuak tribe. Although the majority of the population were formally organized into farmers association, they do not produce cash crops. They live on fishing, hunting and on small scale subsistence farming, growing maize and sorghum. The adult males still travel away from their villages to gold mining fields or coffee

growing areas to make money.

The refugees migrate from different areas of southern Sudan which are known to be endemic for human trypanosomiasis and filariasis. A large number of migrants have occupied the western part of the region, in Itang site along the Baro- Akobo rivers and the second group arrived lately in 1987/88 to the southern part of the Region, at Pingnudo site along the Gilo river.

The Ethiopian settlers arrived to Gambella during 1985/86 resettlement programmes from drought affected Regions of Wollo, Tigray and Shoa.

The study subjects, the indigenous population and the settlers were selected using a simple random sampling method and the refugees using systematic sampling. The list of the head of the house hold including the total population was obtained from the farmers association office in case of the settlers and the indigenous population. The number of house holds required was estimated and then 10% was randomly selected using random table. All members of the selected house holds were included in the study. As of the refugees their way of settlement was not suitable for the above method of sampling. So we used systematic sampling method. The list of the individuals was obtained from the block leaders. All were informed to come to their usual ration dispensing place by the leaders for the examination. 10% of the population was selected using every other 10th individual among the population. For all selected individuals physical examination was done by a physician before blood sample for haemoparasites was taken. All positive cases were treated in the nearby clinics and when found necessary also referred to the Gambella hospital.

Blood sample collection. A total of 875, 1800 and 1400 blood samples were collected and analyzed for haemoparasites from resettlers, indigenous populations and refugees, respectively, using conventional blood films and MHBCT applied for the three study group and M-AECT only for the refugees as it is described here under.

Blood films. Thick and thin blood smears were prepared on the microscope slide following the standard procedure recommended by WHO (8). The smears were allowed to dry and stained with Giemsa and examined for the presence of haemoparasites (trypanosomes, malaria, blood microfilaria) under the microscope at the magnification of x100. In addition to the stained blood smears preparation, wet films were mounted and examined on the spot for all clinically suspected individuals for sleeping sickness infections.

Microhaematocrit Buffy Coat Technique (MHBCT). The MHBCT, also known as Woo's method (9) was performed as modified by WHO (8). Briefly, about 60 µl of blood was taken by a finger prick in heparinized haematocrit capillary tubes in a duplicate from each patient. One end of the capillary tube was sealed with critoseal and centrifuged for 5 minutes at 3000 rpm. Then the capillary tube was attached on a microscope slide with scotch tape. Both Trypanosomes and blood microfilariae were detected in the buffy fraction of the capillary tube under the microscope at the magnification of x10 and then by x40 for confirmation.

Miniature Anion Exchange Centrifugation Technique (M-AECT). This technique was originally developed as a laboratory research tool to detect low parasitaemia of trypanosomes (10). Later on it was designed and prepared in a form of sterile kit, Para- Sight Sterile Kit for field application. It is based on the capacity of cellulose fragments equilibrated by a buffer, Phosphate Saline Buffer with Glucose (PBSG), PH, 8.0 to have a surface static electrical charge. Fresh infected whole blood is allowed to run through a column of equilibrated DE-52 or pre- swollen Diethyl Amino Ethylene Cellulose column (DEAE or DE-52, whatman, England). The blood cells are retained by cellulose while the parasite (trypanosomes and blood microfilariae) are eluted with the buffer as both differ in their electrical charges. The eluate is collected in a special tube centrifuged and the parasites concentrated at the tip of the tube. Then mounted on a viewing chamber and examined under the microscope with the magnification of x10 and x40 for both trypanosomes and blood microfilariae.

Results

Blood microfilariae *Mansonella perstans* were detected in the blood of 75 individuals, that is 1% from indigenous the Anuaks and 4.1% from the Sudanese refugees. The microfilariae of *M.*

perstans distinguished from those of *W.bancrofti*, *Loa loa* and *Burgia* species by the absence of sheath. In addition positive stained blood smears were sent to London School of Hygiene and Tropical Medicine for confirmation. Among the other blood microfilariae cases of *W. bancrofti* was also detected. But the detailed information and result is supposed to be prepared by one of our colleagues (S.A)

During the screening of indigenous population, with Microhaematocrit Buffy Coat Technique (MHBCT) many more microfilaraemic positive cases were detected (1.0%) than with the conventional thin and thick stained blood smears (0.33%). Amongst the refugees Miniature Anion Exchange Centrifugation Technique (M-AECT) detected more than twice (4.1%) as many microfilaraemic cases as MHBCT (1.9%) and more than four times as stained blood smears (1.0%) (Table 1). However, all cases detected by the stained blood smears were also detected by the other two methods. Using wet film mounting, only a single case of microfilaria was detected which also was detected by stained blood films.

The infection was higher in young age group, 15-30 years, (61.1%) and (84.2%) in Anuaks and refugees, respectively and have a significant difference ($P<0.01$) than children 5-14 years and adults above 30 years. No microfilaria was diagnosed in children below 5 years. Higher positivity was observed in males (66.7%) and (94.7%) in Anuaks and refugees, respectively. Regarding the over all positivity, there is a significant difference ($P<0.01$) among the two study groups (Table 2). No blood microfilaria was detected in the blood of resettlers.

Table 1. Determination of *Mansonella perstans* infection rate by Blood films, MHBCT and M-AECT.

Groups	Total Exam.	Blood Thin Pos	film thick% Pos	MHBCT		M- AECT		Total Pos	% Pos
				Pos	% Pos	Pos	% Pos		
Anuaks	1800	6*	0.33	18	1.0	-	-	18	1.0
Refugee	1400	14	1.0	26	1.9	57#	4.1	57	4.1
Settler	875	-	-	-	-	-	-	-	-

* is also detected by MHBCT

includes cases detected by the other two methods

Table 2: Age and Sex distribution of *M. Perstans* infections

Group	Total Pos	Sex	Age				(NO) Pos
			1-4	5-14	15-30	31+	
		M	-	3	7	2	12
				16.7%	38.9%	11.1%	66.7%
Anuaks	18	F	-	1	4	1	6
				5.6%	22.2%	5.6%	33.3%
N=1800		Total	-	4	11	3	18
				22.2%	61.1%	16.7%	100%
		M	-	4	47	3	54
				7.0%	82.5%	5.3%	94.7%
Refugee	57	F	-	2	1	-	3
				3.5%	1.8%	-	5.3%
N=1400		Total	-	6	48	3	57
				10.5%	84.2%	5.3%	100%

Discussion

During a sleeping sickness survey in 1989-92 in Gambella, South Western Ethiopia, microfilariae of *Mansonella Perstans* were detected in the blood of the indigenous population, the anuaks (1%) and the refugees from Southern Sudan (4.1%). No blood microfilarial infection was observed in the resettlers.

The infection rate is higher in young age groups, 15-30 years 84.2% in the refugees and 61.1% in Anuaks than the children below 15 years and older age groups. Eventhough it is not statistically significant there is some differences in positivity in males than females in both study subjects. This could be attributed to the occupational division and the biting nature of the vector (6). Males visit forests more frequently than females, and the culicoides bite mostly in dense patchy forests than open fields.

The pathology ascribed to *M. perstans* infection is not well recognized nor is the clinical symptomology well recorded (12). It was of interest to us therefore that many of the individuals who had high microfilariae count commonly complained of joint and back pains, headache, weakness, pruritus, etc. Did sleeping sickness whose symptoms are well known suggests them (8) or were they due solely to *M. perstans* infections regardless of the co-existence of malaria infection in the area. While we can not speculate, it is better to take into account that even an infection which normally might cause little or no illness could become serious in combination with other intercurrent diseases.

Previous studies indicated that there was a prevalence of 24% *Wuchereria bancrofti* infections among the Anuaks in the area (3). In the present study cases of *W. bancrofti* were detected among the indigenous population, the Anuaks (NRHI, unpublished report). That is the microfilariae of *W. bancrofti* was identified from stained blood smears by the presence of a sheath and subterminal arrangement of the nuclei.

The interesting observation made in the present study is that, M-AECT detect almost more than twice as many microfilaraemic cases as MHBCT and more than four times by stained blood smears among refugees which is quite in agreement with Lumsden et.al. and others (10,11). Due to the shortage of the kit we could not apply M-AECT, during the screening of the indigenous population the Anuaks and the resettlers.

Though M-AECT is the most sensitive test known so far, it needs further improvement in field conditions. However, the preparation of sterile kit of optimal size under difficult conditions avoids the need for refrigeration (8,11). Thus sterile kits are stable, and always the sterility is needed to prevent faulty diagnosis due to the presence of other motile micro-organisms in the column. As compared to M-AECT, MHBCT is relatively less sophisticated and could therefore easy to manipulate in the field.

Based on these facts, the complete epidemiological profile of lymphatic filariasis and other blood filariasis in the area, including periodicity, density of the microfilaria and clinical picture of the disease should be studied. By large, as compared to M-AECT, MHBCT with its relative merits of low sophistication, could therefore, find application in some blood filariasis survey. In addition application of standard diagnostic techniques such as Millepore filtration methods, Knott's concentration technique (12,13), etc. for blood filariasis examination is recommendable.

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Original article

The Attitudes of students, parents and teachers towards the promotion and provision of condoms for adolescents in Addis Ababa

Abstract: A cross-sectional descriptive survey through a self-administered, anonymous and structured questionnaire was conducted from September to December, 1993, in ten high schools in Addis Ababa to determine the sexual behaviour of adolescents, their knowledge about AIDS, attitudes and practices regarding condoms, their attitudes towards the promotion and distribution of condoms in schools, and towards the incorporation of health and sex education into the regular teaching curricula and into the teachers' training curricula.

A total of 910 parents, 755 students and 232 teachers participated in the survey. The results showed that, of the 755 students, 39.8% of the boys and 5.6% of the girls have had sexual experience. Peer pressure (35.2%) and force (21.6%) were the most important factors that precipitated the first sexual encounter. Ten percent of the students had coital contact with a commercial sex worker. Only 42.2% of the sexually active students used condoms on their first sexual encounter, and only 27.7% used condoms continuously on their subsequent sexual encounters.

An overwhelming majority in each of the three categories, (92.6% of the students, 98.7% of the parents and 96.1% of the teachers), approved the incorporation of health education into the regular teaching curricula. The idea of sex education in schools was also approved by 80.1% of the students, 90.9% of the parents and 96.1% of the teachers.

This study, based upon the findings, recommends that education and health policy makers make relentless effort to commence health and sex education by incorporating them into the regular curricula; the implementation of subsequent surveys to identify the most effective and acceptable routes of condom distribution in school; and implementation of similar surveys in the rural settings to assess the attitudes in a different setting and acquire a more general overview for the whole country. [*Ethiop. J. Health Dev.* 1997;11(1):7-16]

Introduction

Since its identification in 1981, AIDS has been spreading over the world at an alarming speed. By 1990, it was estimated that there were 8-10 million people infected with HIV (1). This figure was expected to reach 15 million, that is 7.4 million males, 5.9 million females and 1.3 million born with infection, world wide by 1993 (2). In Africa alone more than 3000 new infections occur every year, and if uninterrupted, this figure is expected to reach 30-40 million by the year 2000 (3). Characteristically 20% of those infected with HIV are expected to be between the ages of 20-29 (4). Given the long incubation period, with the mean latency time of the illness (eight years) (5), a substantial number of those infected may have acquired the disease at earlier ages, perhaps in their adolescence.

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Reports on the situation in Ethiopia demonstrate that, from January 1985 to June 1993, 6726 AIDS cases were reported from 47 hospitals. Forty one percent of these were residents of Addis Ababa. The largest reported number was in 1992, accounting for 48.41% of the total reported since 1986. By 1993 the sex ratio of those reported was 1.6:1, male to female, and the 15-19 age group accounted for 5.81% of the reported cases, while those aged 20-24 years constituted 19.81% of the total cases. About four percent of the total reported cases were students (6).

The Ethiopian National AIDS Control Programme (NACP) estimated that by the end of 1994, 536,600 persons will carry the disease, and in the same year 18,315 new cases of AIDS will be diagnosed (7).

All the above reports clearly point firstly to the rapid spread of the disease, and secondly that adolescents, the focus of this study, represent quite a significant number of those being predisposed to and even acquiring this dreadful disease.

In many countries (particularly the developing ones), adolescents account for a larger proportion,

around 20-25% of the total population (8). Constituting a large proportion of the population and also from various behavioural, cognitive and developmental perspectives, adolescents are labelled as a vulnerable group and deserve attention in terms of research and prevention.

Many studies have been conducted all over the World to assess the sexual behaviours of adolescents. All these studies claim that adolescents are in fact sexually active, with figures ranging from 17.3% to 83% and that this trend is increasing. Due to all these, adolescent sexuality is not an issue to be ignored (9-20, 22).

The very few studies conducted in Ethiopia also demonstrate a similar pattern. Alarming, most of the above studies including those conducted in Ethiopia (8, 23, 24, 25, 26), demonstrate that those sexually active adolescents do not practice safe sex.

The fact that condoms significantly reduce the risk of HIV infection is beyond discussion (27). With the AIDS crisis and increase in other STDs and the need to develop safer and more effective barrier contraceptives, attention has been reoriented to the only available protection, the condom (28). This being the case, we can no longer ignore the association of condoms with adolescents. We cannot also rely only on talking about abstinence (though this should be promoted also). The facts will force us to reorient our efforts towards educating them to exercise safe sex, through maintaining a single partner, and if this is not possible, to always use condoms. This view has also been supported by many investigations (9, 11, 16-21, 24, 25-28).

There are two concerns that need to be addressed at this juncture. The first one is, if condoms are to be distributed to students, will this be promoting their sexuality? And secondly, is this really what they want? Perhaps the strongest and most conclusive rejection of speculations such as this was provided by the WHO (1993). It critically reviewed 19 studies conducted all over the world and disproved that sex education and availability of contraception encouraged sexual experimentation. In fact, sex/AIDS education encouraged adolescents to delay sex initiation and to practise safe sex if sexually active. It also found out that schools that promoted postponement of sex and use of condoms were more effective than those which promoted abstinence alone (29).

The above being the case elsewhere, this study tried to assess the sexual activity pattern of high school students in Addis Ababa viz-a vis their parents and teachers attitudes towards the issue of condom promotion and provision of health education in schools.

Methods

A cross-sectional survey on sexual activity patterns, attitudes and knowledge regarding the use of condoms, attitudes towards the provision of condoms at schools, and attitudes towards incorporating health and sex education into school curricula was conducted among students, parents and teachers in ten high schools in Addis Ababa over a period of one month (December, 1993).

The source population consisted of students, parents and teachers of all 38 high schools in Addis Ababa. The study population was obtained from ten high schools that were selected using the probability proportionate to the size (PPS) method. Students of both sexes attending grades 9-12, parents and foster parents of students in the selected schools, and teachers teaching in the same high schools were considered to be eligible for the study.

A multistage sampling procedure was used to select subjects to be included in the study.

Two structured and pretested questionnaires were developed in such a way that measurement of the following variables was attained: The sexual activity patterns of high school students, including (a) the proportion of sexually active students; and (b) the presence of any high risk behaviours, such as multi-partner sexual contact and the use/non-use of condoms; attitudinal variables (a) on the knowledge and attitudes of students regarding AIDS and condoms; (b) on the attitudes of students, parents and teachers towards the promotion and provision of condoms in high schools; and (c) on the attitudes of students, parents and teachers towards the provision of health and sex education as a separate course in high schools.

Other socio-demographic variables, such as age, sex, parental marital status, monthly income, religion and ethnic grouping were included to allow later determination on how they relate to the

major dependent variables. All data were collected using trained twelfth grade data collectors and nurse supervisors.

Data were then analyzed using the EPI-INFO (30) and SAS programmes.

Results

A total of 755 (89%) students, 910 (91%) parents and 232 (92%) teachers participated in the study. The mean age of the students was 14.74 years (SD 5.13). The youngest participant was 12 years old and the oldest was 23 years. Almost an equal number of boys and girls participated in the study, 359 (47.5%) and 372 (49.3%), respectively. Christianity was the predominant religion (85.9%). Most of the students were Amhara (56.4%). Twenty eight percent of the students were ninth graders, 24.6% tenth graders 25.7% eleventh graders, and 20.1% twelfth graders.

Regarding the parents, the mean age of the participants was 42.25 years (SD 10.88). The youngest respondent was 20 years and the oldest 104 years. Five hundred forty three (59.7%) of the parents were females. Eighty five percent were Christians and 51.1% were Amahara. Seventy two percent of the parents were married. Fifteen percent were illiterate.

The mean age of the teachers who participated in the study was 37.4 years (SD 10.7). The youngest participant was 23 and the oldest 58 years old. One hundred ninety six (84.5%) of the respondents were males and 33 (14.2%) were females. Here also, Christianity was the predominant religion accounting for 90.1%. Fifty two percent of the teachers were Amhara whereas 64.7% of the teachers were married (Table 1).

Out of the 755 students respondent, 148 (19.5%) admitted to having had a coital experience at least once prior to this study, of which 124 (83.8%) were boys 20 (13.5%) were girls, accounting for 39.8% of the boys and 5.6% of the girls. The earliest reported age of onset of sexual intercourse for girls was fourteen years with mean age of onset being 15.30 years (SD 5.39). The earliest age of commencement of sexual activity for the boys was twelve years with mean age of onset being 16.45 years (SD 4.02).

Peer pressure was the frequently reported factor that led to the first sexual encounter, accounting for 35.2% of the sexually active respondents, followed by being forced (21.6%), alcohol (11.5%) and drugs (10.3%). Twenty seven percent admitted that they first performed sex in hotels, followed by partners' house 20.2%, and own house 20.2%. Hotels are also the most frequent places where the sexually active students had sex after the first sexual encounter, accounting for 29.1% of the responses.

Table 1: Social and demographic characteristics of students, parents and teachers who participated in the survey (Addis Ababa, December, 1993)

Socio-demographic characteristics	Students No. (%) (n=755)	Parents No. (%) (n=910)	Teachers No. (%) (n=230)
Age			
10 - 12	2 (0.3)	-	-
13 - 14	74 (0.8)	-	-
15 - 16	280 (37.1)	-	-
17 - 18	279 (37.0)	-	-
19 - 20	42 (5.5)	-	-
no response	76 (10.1)	-	-
20 - 30	-	119 (13.1)	15 (6.5)
31 - 40	-	354 (38.9)	115 (49.6)
41 - 50	-	282 (31.0)	80 (34.5)
51 - 70	-	143 (15.7)	10 (4.3)
71 - 90	-	11 (1.2)	-
>90	-	1 (0.1)	-
no response	-	119 (13.1)	12 (5.1)
Sex			
males	359 (47.5)	367 (40.3)	96 (41.7)
females	372 (49.3)	543 (59.7)	33 (14.2)
no response	24 (3.2)	-	3 (1.3)
Religion			
Orthodox Christian	629 (83.3)	780 (85.7)	192 (83.5)
Muslim	52 (6.9)	70 (7.7)	12 (5.2)
Protestant	45 (6.1)	26 (2.9)	14 (6.0)

Catholic	13 (1.7)	25 (2.7)	5 (2.2)
Other	9 (1.2)	9 (1.0)	5 (2.2)
No response	6 (0.8)	-	4 (1.7)
Ethnic group			
Amhara	426 (56.4)	465 (51.1)	121 (52.2)
Oromo	128 (17.0)	177 (19.5)	35 (15.1)
Tigian	73 (9.7)	98 (10.8)	26 (11.2)
Gurage	74 (9.8)	94 (10.3)	17 (7.3)
Other	40 (5.3)	76 (8.3)	29 (12.5)
No response	14 (1.8)	-	4 (1.7)
Marital status			
Married	533 (70.6)**	657 (72.2)	150 (64.7)
Never married	22 (2.9)	107 (11.8)	66 (28.4)
Separated	12 (1.6)	-	-
Widowed	123 (16.3)	2 (0.2)	2 (0.9)
Divorced	52 (6.9)	92 (10.1)	12 (5.2)
No response	13 (1.7)	52 (5.7)	2 (0.8)
Family income (birr/month)			
0 - 100	-	345 (37.9)	-
101 - 200	-	203 (22.3)	1 (0.4)
201 - 400	-	198 (21.8)	5 (2.2)
401 - 800	-	98 (10.8)	175 (75.4)
801 - 1200	-	35 (3.8)	39 (16.8)
>1200	-	31 (3.4)	8 (3.4)
no response	-	-	4 (1.7)
Educational level			
9	211 (27.9)	-	-
10	186(24.6)	-	-
11	194 (25.7)	-	-
12	151 (20.1)	-	-
no response	13 (1.7)	-	-
Not educated	-	144 (15.8)	-
Elementary incomplete	-	239 (26.3)	-
Elementary complete	-	109 (12.0)	-
Socondary incomplete	-	71 (7.8)	-
Secondary complete	-	169 (18.6)	4 (1.7)
University incomplete	-	29 (3.2)	82 (35.3)
University complete	-	51 (5.6)	138 (59.5)
Other	-	98 (10.8)	7 (3.0)
No response	-	-	1 (0.4)

N.B. * Socio demographic characteristics ** Parents of students

Regarding the duration of acquaintance with the first sexual partner, 14.9% were acquainted only for hours, 3.0% for less than one week, 18.2% for two or more weeks, 20.9% for many months, and 44.6% for many years.

Ten percent of the sexually active male students admitted having sex with commercial sex workers. Of the respondents, 37.2% never changed sexual partner since their first encounter, followed by 18.2% once, 18.2% twice, 5.4% three times, and 12.2% four or more. Sixty one percent of the sexually active students admitted to having had sex within the last three months.

Only 43.2% of the sexually active students knew about condoms on their first coital encounter. Eighty two percent of those did not use condoms on their first sexual encounter. Only 27.7% of the sexually active students claimed that they had continuously used condoms (Table 2).

Fifty two percent of the parents thought that most high school students are sexually active. A substantial proportion of the parents (40.9%) said that they knew nothing about high school students' sexual behaviours. Also on this issue, 81.5% of the teachers thought that most students are sexually active (Table 2).

Forty seven percent of the students felt that the majority of the students do not have enough knowledge about AIDS. Only 38.1% of the parents on the other hand felt that a majority of students do not have enough knowledge about AIDS. More than half of the teachers (52.2%) thought that most students do not have adequate knowledge about AIDS.

Regarding the source of information about AIDS for students, the majority (64.1% of the students, 56.2% of the parents and 52.2% of the teachers) acknowledged mass media to be the most important source of information about AIDS.

When asked about the overall students' knowledge of condoms, 42.4% of the students thought that most students do not know what a condom is, while only 23.8% of the parents and 28.9% of the teachers felt that the majority do not know about condoms. Further more, only 42.8% of the

students, 38.8% of parents and 34.9% of teachers felt that most students know how to use condom properly.

Table 2: Distribution of knowledge and behavioural factors among sexually active students in Addis Ababa, 1993.

Behavioral factors	Students No. (%)
*Factors which led to the First sexual encounter	
Peer pressure	52 (35.2)
Being forced	32 (21.6)
Alcohol	17 (11.5)
Drug	16 (10.8)
No response	31 (20.9)
*Knowledge of condom prior to the first sexual encounter	
Yes	64 (43.2)
No	66 (44.6)
No response	18 (12.2)
*Use of condom on the first sexual encounter	
Yes	26 (17.6)
No	103 (69.6)
No response	19 (12.8)
*Continuous use of condom on subsequent encounters	
Yes	41 (27.7)
No	81 (54.7)
No response	26 (17.6)
*Frequency of sexual encounter during the last 3 months	
None	59 (39.9)
Once	33 (22.3)
Twice	15 (10.1)
Three times	9 (6.1)
Four or more	6 (4.1)
No response	26 (17.5)
*Last sexual encounter	
1 - 5 days	18 (12.3)
1 - 2 weeks	14 (9.5)
3 - 4 weeks	16 (10.8)
1 - 3 months	15 (10.2)
More than 3 months	59 (39.7)
No response	26 (17.5)

* Behavioral actors

Some of the reasons that the respondents felt prevent sexually active students from using condoms include the following: for students the two outstanding reasons were negligence (28.2%) and embarrassment in buying from the shops or pharmacy (26.4%); for parents, fear to buy from shops and pharmacies (30.0%) followed by lack of information and knowledge (23.8%); and for teachers, lack of information and knowledge (24.1%) followed by fear to buy from pharmacies or shops.

Most students (53.6%), parents (70.2%) and teachers (56.5%) said that mass media is the most important source of information about condoms followed by health professionals.

Distribution of condom in schools was approved by 61.6% of the students, 60.0% of the parents and 61.6% of the teachers with 47.0% of the students, 35.4% of the parents and 44.0% of the teachers preferring that condoms be distributed to students freely, while 20.5% of the students, 23.8% of the parents and 31.5% of the teachers chose a very cheap price (Figure 1).

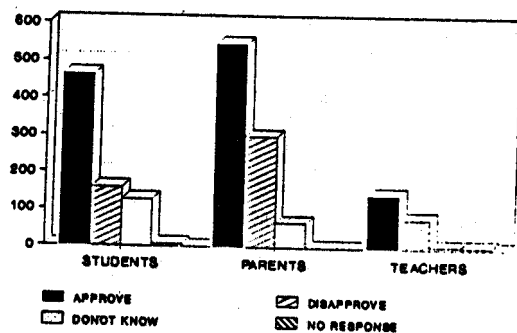


Figure 1: Attitude towards condom provision in schools, Addis Ababa, 1993.

When asked who should distribute condoms in schools, 37.2% of the students chose primarily AIDS Club members (students) followed by school clinics (23.8%). Parents, on the other hand, preferred school clinics as their first choice (24.3%), followed by school guidance officers (19.2%). Teachers preferred school guidance officers (24.6%) followed by school clinics (18.5%).

A stratified analysis that was conducted to measure the effect of the various socio-demographic variables on the attitudes towards distributing condoms in schools revealed only age for parents (the older generation tending to disapprove), sex of students (females tending to disapprove), and religion for teachers (Muslims tending to disapprove), to have a statistically significant influence upon the particular attitude taken by the different groups. This was found to be the case both before and after controlling for the other factors.

The majority of the students (92.6%), parents (98.7%) and teachers (96.1%) thought that there is an urgent need to start health education in schools. Forty one percent of the parents and the 40.9% teachers preferred that it should start at the elementary level (1 - 5) while 46.8% of the students chose the secondary level (9-12). As to how it should be provided, 37.0% of the students, 57.4% of the parents, and 53.0% of the teachers thought that it should be given as a separate course with its own curriculum (Figure 2).

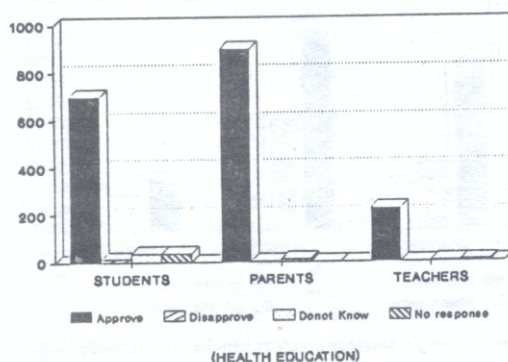


Figure 2: Attitudes towards health and sex education in schools, Addis Ababa 1993.

Considering sex education, the majority in all the groups, students (80.1%), parents (90.9%), and teachers (96.1%) felt that sex education should also be provided in schools commencing in the junior secondary grades (7-8).

Discussion

This study gives important information regarding the sexual behaviour of students in Addis Ababa, their high risk behaviour, and possible protective measures that could be implemented in an effort to help control the spread of AIDS among adolescents.

Nineteen percent of the participating students admitted to having sexual experience accounting

for 39.8% of the boys and 5.6% of the girls. These figures are relatively lower when compared with the results of similar studies. In U.S.A and Canada the figures were between 46% - 72% for boys and 24% - 72% for girls (9, 10, 11, 12, 31); in Europe it was between 18.9% - 78% for boys, and 17% - 45% for girls (15, 16, 17); in Latin America 42% - 78% for males, and 12% - 55% for females (14); in Africa the figures in general were above 30% (18, 19, 20, 21, 22); in Ethiopia in Harar 20% for females, 65% for males (8), in Jimma 40% for both sexes (23), Northwest Ethiopia 52.8% for both sexes (24) and in Addis Ababa 53% for boys and 24% for girls (25).

The result for the boys was fairly comparable to most of the previous findings. But when compared with the other studies the girls' data appeared to be relatively low. This could probably be due to the cultural influences, which, despite the strong assurance of anonymity, the girls did not provide their true response, or, it may be due to boys being more sexually active. The fact that boys are more sexually active than girls has been demonstrated by most of the above studies. Also, that there is a tendency for the girls to persistently under-report their sexual behaviour has again been confirmed by some of the studies (11, 15). This may have also been the case in this study.

Seventy eight percent of the sexually active respondents claimed that their first intercourse was initiated by either peer pressure (35.2%), being forced (21.6%), alcohol (11.5%) or drugs (10.3%) indicating either unplanned encounters or circumstances creating unfavourable situations for making responsible decisions.

Thirty six percent of the sexually active students had had very minimal acquaintance (less than one month in most cases) with the person that they had sex with. An important majority of them (10.1%) admitted to having had sex with a commercial sex worker. Fifty four percent of the sexually active students reported also that they had had multiple sexual partners. All the above findings clearly and alarmingly indicate to a prevalence of high risk behaviour. This has also been the finding of various other previous investigations (10, 12, 25).

Another characteristic feature which makes adolescent sexual activity high risk is their either non or very minimal use of any protective measure, specifically the use of a condom. Only 17.6% of the sexually active students used condoms in their first sexual encounter, and only 27.7% claimed to have used a condom continuously on their subsequent sexual encounters. Similar studies conducted in Kenya, Nigeria and U.S.A also support this finding (8, 12, 18, 18, 21). Although condoms are available in many places, a substantial number of the sexually active adolescents claimed that they did not know about condoms when they had their first sexual encounter (44.6%). This indicates that information pertaining to condoms is not provided to adolescents early enough, that is when they are at the period of initiation of sexual activities. This was also the attitude of a substantial proportion of students (42.4%) who thought that most students do not have adequate information about condoms and AIDS (47.2%).

This study, therefore, demonstrates that students are sexually active, and they continue to practise risky behaviours due to either lack of adequate information related to condoms and AIDS or barriers preventing them from easily acquiring condoms when they need them. To improve on this situation two possible intervention measures are possible: bringing about behavioural changes such as exercising abstinence or, if this is not possible, pushing them to remain steadfast with only one partner, through education, and make condoms more accessible and through acceptable routes.

Schools are the places where students acquire most of their knowledge. The role schools could play to bring about desired behavioural changes among their pupils is also potentially great. In schools, students are provided with information on various aspects of life through well organized and developed instruction methods. This makes schools ideal places to commence and continue educating them on aspects of life. Since issues related to health are also one aspect of life, these should be given due attention and emphasis in the school educational environment if an overall desirable behavioural change is to be achieved among the learners.

The role schools have played in the provision of information and education on matters related to health has, up to now, been very minimal or nonexistent. This is evident in this study, that mass media rather than schools are more important source of information followed by health professionals. Very few students, parents and teachers considered schools to be important sources

of information for adolescents.

Gebre, (1990) also came up with similar findings in Addis Ababa, in that 63.8% of the students chose mass media to be the best source of information for the same subject followed by health institutions and professionals (25). In his survey, only 10.3% chose schools to be the best sources of information. Other studies conducted elsewhere, on the other hand, indicate that schools rather than mass media to be the major source of information. For example, in Kenya, where there is an elaborate Health Education in schools, students rely more on schools as their most important source of information on AIDS and related matters than mass media. This indicates the potential that schools have in becoming important and reliable sources of information for students on issues related to sexuality and AIDS (19, 32).

It is obvious that information provided by the mass media is targeted at the general public and does not particularly address adolescents. It is also fragmented and lacks continuity, not all issues pertaining to sexuality, STDs and adolescence are emphasized. In fact students may not understand the message, especially in their earlier years when they are conceptually not well versed on these issues, but in reality there is an urgent need to start the education before they actually become sexually active and risky behaviours become well established.

This is the reason why almost all of the students, parents and teachers look to schools to assume an increased role in the provision of health education for adolescents. The vast majority of students, parents and teachers felt that there is a need to commence health education immediately. Moreover, most of the teachers and the parents preferred that health education start at the elementary level, while most of the students felt that it is better to start it in high schools, moreover most parents, teachers and students also felt that it is better if health education is provided in schools as a separate course.

Teachers and similarly, most students wanted health professionals to assume an important role in the provision of health education in their schools. A vast majority of the teachers also approved the incorporation of health and sex education in the teacher's training curricula.

Regarding sex education, the majority of the students (80.1%), parents (90.9%) and teachers (96.1%) felt that there is a need to start sex education. Parents (42.0%) and teachers (47.8%) felt that junior secondary grades (7-8) are better places to start sex education, while students (53.8%) preferred secondary grades.

This study, therefore, has quite strongly demonstrated that there is a strong desire by the participants for health and sex education to start in schools, with many of the parents and teachers desiring that it starts even at earlier grades.

Besides the provision of education another equally important intervention possibility that needs to be given similar attention is the provision of condoms in schools. This is not only to protect students from acquiring AIDS but also from other STDs and unwanted pregnancies.

It has been described in detail above that despite their availability elsewhere, sexually active adolescents do not use condoms as hoped. This obviously led us to look for more accessible and effective ways of providing them, like in the schools.

This is because at least a substantial proportion of adolescents are present in schools particularly in urban settings. Therefore, if condoms are made accessible to them in schools, this may relieve them from the fear of buying condoms from other exposing places, which was shown in this study to be one of the major obstacles.

Another equally important issue that should be given due attention, if increased condom provision is anticipated, is the presupposition that the public may fear that it might promote promiscuity and therefore not accept this idea. No study has substantiated this, and in fact, most studies conducted in this area have recommended the provision of protective measures. This is also what is strongly recommended by the WHO (13, 17, 26, 28, 29, 31).

The findings in this study might come as a surprise. But, in a strongly cultural, religiously dominated and more traditional country, coming up with such a finding may suggest the following: first, it may indicate that the population is becoming more aware of the consequences of AIDS and is gradually accepting whatever measures it has to be taken to stop the spread even if it is entirely

against its religious and/or traditional beliefs; and secondly, it may also suggest that modernization is gradually eroding the traditional cultural values. On the other hand since this study was conducted in an urban setting, where the population appears to be culturally liberal, it may not reflect the true feelings of the general population.

A change in the traditional values for parents is also suggested by the finding in this study through stratified analysis that indicated a decreasing trend to accept this idea as age increases.

This study has demonstrated that many adolescents students in Addis Ababa high schools are sexually active. Not only are they sexually active, but they exercise risky sexual behaviours, despite large mass media campaigns about AIDS and its consequences and the availability of condoms.

Most students, parents and teachers were not found to be fully confident of students' knowledge about AIDS and condoms. An overwhelming majority of the students, parents and teachers supported the idea of immediate commencement of health and sex education in schools. A substantial proportion of them also approved the promotion and distribution of condoms in high schools.

Therefore the conclusion generally is that the population in general and policy makers in particular could no longer continue to ignore the issue of steadily growing and alarming risky sexual behaviour of adolescents. This, therefore, indicates for an urgent implementation of comprehensive education and prevention programmes.

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Community-based Family Planning services: A performance assessment of the Jimma FP CBD project

Antenane Korra¹

Abstract: Community Based Distribution (CBD) programs reach beyond the clinic walls to provide contraceptives in the communities where people live. The ultimate goal of CBD programs is to improve the accessibility of contraceptive services and supplies. In view of this, the Family Guidance Association of Ethiopia (FGAE) has recently initiated CBD programs in some rural areas. The aim of this study is to assess the overall performance of the Jimma CBD project with respect to achieving the set objectives and measure the changes that have occurred in the levels of knowledge and use of family planning methods among the eligible couples. Results of the study showed, that the knowledge of women about contraception appeared to be higher (76.9%) than the proportion reported in the baseline survey (36.9%). Comparing the contraceptive prevalence rate of 1.3% reported in the baseline survey, the rate reported in this study is found to be substantial (15.9%). Generally, the project achievement enlisted in the period of 21 months of operation was found to be impressive. Thus, replication of the program in other rural areas should be sought. [*Ethiop. J. Health Dev.* 1997;11(1):17-22]

Background

In developing countries, family planning services are delivered mainly through health institutions. In the African context, the majority of the population does not have similar privileges of ease of access to facilities for family planning service delivery, especially in rural areas. Though in some countries clinic-based family planning service delivery facilities exist, these services are far from being adequate to meet the needs of the population demanding such services, (1).

The introduction of modern family planning services in Ethiopia dates back to 1966, when the Family Guidance Association of Ethiopia (FGAE) was founded as a non-governmental and not-for-profit organization to provide information, counselling and clinical services to families who voluntarily express their need and desire for spacing the birth of their children.

when the Ministry of Health integrated family planning with the Maternal and Child Health care (MCH) services in 1980. Since then, family planning services have been offered through government static health institutions and other organizations. This approach, which has been clinic-based and biased towards curative medicine, has remained remote from the potential and large segment of family planning users residing in rural areas.

To make family planning information and services accessible and affordable to the hard- to-reach rural communities, community-based distribution (CBD) programs have been practiced in many developing countries and this approach brought about successful results in Asian and African countries. In view of this fact, expansion of community-based distribution of family planning services is indicated as one of the basic implementation strategies in the National Population Policy of Ethiopia launched in July 1993, (2).

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The services have been further strengthened

"The Jimma Family Planning Community- Based Distribution Project", funded by Population

Concern (UK), is serving rural communities of "Mana and Kersa" woredas of Jimma zone, around Jimma town, South West of Ethiopia. The project is designed to expand family planning services through increased knowledge of family planning concept by promoting accepted contraceptive mix with community participation. Specifically, the project is aiming at increasing knowledge of family planning among the couples from the observed 36.9% to 75% and raising the contraceptive prevalence rate from 1.3% to 15% in the period of three years (3).

With these specific objectives the project had been in operation for about 21 months. Thus, this mid term review has become imperative to assess its achievements with respect to attaining the intended objectives and to identify the major constraints that hinder project performance.

This study tries to assess the overall performance of the Jimma CBD program during the 21 months it has been in operation, and to identify the strong and weak points of the program.

Specifically, the objectives are to:-

- i) measure and compare the level of knowledge and use of family planning of the eligible couples with that of the baseline survey conducted prior to the program of intervention;
- ii) assess to what extent the objectives of the project have been achieved.

Methods

Data for the analysis have come from three sources. A survey was carried out in April, 1994 to measure the levels of knowledge and practices of family planning of the community. It was undertaken in five randomly selected project sites, namely, Doyo Awaso, Gerukie Jimatie, Kitimbilie, Somodo Abujedi and Kersa Balto. These sites were the areas where the baseline survey of the project was carried out.

It is, however, worth to mention the differences in sampling methodologies of the two surveys. For the baseline, data were collected from all eligible women residing in the five project sites, while a sample of eligible couples have been randomly selected for this study. To this effect, it was planned to interview a sample of 225 couples, and about 98% of the intended respondents were successfully interviewed. The sample size was determined with critical considerations of cost and logistical feasibility. Thus, 10% of the eligible couples residing in the selected areas were taken. Samples were allocated to each survey site using probability proportional to size, proportion being the size of total eligible couples in each site. A Systematic sampling technique was then employed to select couples for interview and structured questionnaires were administered by enumerators. Service providers (CHAs) were also interviewed and further, records of the project were reviewed.

Results

The data on background characteristics of the interviewed respondents indicate that women in the age of 25 to 29 years constitute

Table 1: **Percentage distribution of Respondents Knowledge about Contraceptives by Method, 1994**

Methods	Mid-Term		Baseline Female (1447)
	Female (216)	Male (197)	
Pills	95.1(158)	91.2(125)	95.1(508)
Condom	13.8(23)	49.6(68)	13.8(74)
Foam Tablet	7.2(12)	8.7(12)	7.5(40)
Injectable	13.8(23)	19.7(27)	25.8(138)
IUD	4.8(8)	4.4(6)	15.3(82)
Total	76.9(166)	69.5(137)	36.9(534)

N.B. Figures in parenthesis are number of cases

Table 2: **Distribution of Respondents Reporting Ever Use of Contraceptives by Method, 1994**

Methods	Mid-Term		Baseline Female (1409)
	Female(220)	Male(210)	
Pills	96.0(48)	71.4(25)	96.8(60)

Condom	4.0(2)	22.8(8)	-
Injectable	2.0(1)	2.9(1)	-
IUD	-	-	3.2(2)
Total	22.7(50)	16.7(35)	4.4(62)

N.B. Figures in parenthesis are number of cases

the largest portion than women of other age groups. For the case of interviewed men, however, most are found at the age of above 50 years. With the mean age of 32.4 years, women are found to be younger by about 8 years than men. Further, respondents of the study area are predominantly muslim with low educational attainment. Of the total interviewed women, about 73% are illiterate and this proportion is small for the case of men.

The study also showed that the areas under investigation are characterized by high fertility. The reported total fertility rate (TFR) is estimated at 10.9 children per woman and both women and men want to have about three additional children.

Knowledge of respondents about health posts (service delivery points of the project) showed that women knew and visited the health posts more than men. In conjunction with this, the study revealed that the knowledge of women about contraception appeared to be higher (76.9%) than the proportion reported in the baseline survey (36.9%) (4). A substantial number of them (about 95%) have heard about the pills. The level of men's knowledge about contraception is also found to be high with 69% of them reporting that they have heard at least one method of contraception (Table 1). A good number of women (72.1%) and men (65.5%) stated that their sources of information were the CBD agents.

With regard to the use of contraception, the study showed that a significantly higher proportion of ever use (22.7%) is reported among women than ever use reported in the baseline survey (4.4%), (4) and a considerable number of them have used pills (Table 2).

As to the level of current use, about 16% of women and 13% of men were using contraceptive method at the time of the survey. Comparing the contraceptive prevalence rate of 1.3% reported in the baseline survey, (4) the rate reported in this study is noticeably high. As expected, the prevalence of pills is high, about 94.4% of current users were using the method (Table 3) and health posts are mentioned by almost all ever users (about 94%) as the main source of contraceptive supply.

Table 3: **Distribution of Current contraceptive Users by Method, 1994**

Methods	Mid-Term		Baseline Female (1338)
	Female(220)	Male(220)	
Pills	94.4(38)	89.3(25)	90.0(18)
Condom	-	3.6(1)	-
Injectable	2.8(1)	3.6(1)	-
IUD	2.8(1)	3.6(1)	-
Total	-	-	10.0(2)

N.B. Figures in parenthesis are number of cases

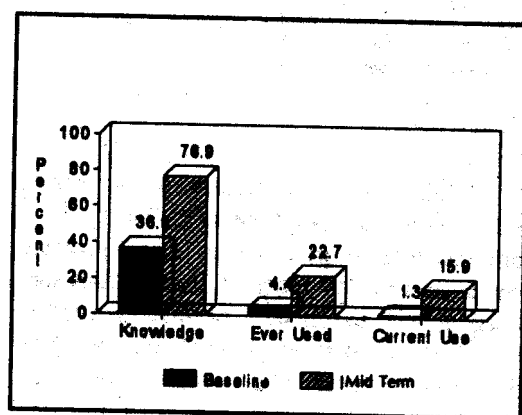


Figure 1: Percentage of Women heard about, ever used and current use of FP method

It is also observed that, among non-users, desire for more children is frequently mentioned as the reason for not practising contraception. In the baseline survey, however, lack of knowledge about family planning was the most frequently mentioned reason for not practising contraceptives (Table 4).

The analysis further revealed that both the CBD agents and the family planning educator assigned to the project, jointly motivated, on the average, 6,700 people residing in all CBD. Accordingly, teaching aid materials (2510 leaflets, 710 booklets and 2346 posters) dealing with different family planning messages have been distributed to support the motivational endeavors.

Table 4: Distribution of Never Users by Reasons for Not Using Contraceptives, 1994

Reasons	Mid Term		Baseline
	Female (161)	Male (163)	Female (1347)
Naturally Spaced	13.6(22)	9.1(15)	-
Desire for more children	37.0(60)	50.6(83)	19.3(260)
Do not want to use	19.8(32)	17.1(28)	0.5(7)
Infertility	14.2(23)	10.4(17)	8.8(119)
Lack of Knowledge about FP	11.7(19)	11.0(18)	67.5(910)
Husband's Opposition	1.2(2)	-	11.3(152)
Health Concern	1.9(3)	0.6(1)	4.9(66)
Religious Reason	-	0.6(1)	3.5(47)

N.B. Figures in parenthesis are number of cases

Table 5: Contraceptives Issued and CYP Generated, July 1992-March 1994

Contraceptive Method	Unitq	Issued	CYP Generated
Pills	Cycles	11277	867
Condom	10955	110	
Foam Tablet	119	24	
Total			1001

Additionally, the records of the project indicated that a total of about 11,300 cycles of different types of pills, about 11,000 pieces of condoms and 119 tubes of foam tablets have been issued to clients. In this connection, Couple Year of Protection (CYP), one of the measures of family planning program output, (5) is calculated, and found that a total of 1001

Table 6: Percentage of Contraceptive Acceptors by Method, July 1992-March 1994

Method	Acceptors	
	New	Revisit
Pills	69.6(1348)	96.0(9523)

Condom	27.4(530)	3.6(<358)
Foam Tablet	3.0(< 58)	0.4(<41)
Total	100.0(1936)	100.0(9922)

NB Figures in parenthesis are number of cases

CYP was achieved within the period of 21 months (Table 5). In other words, within a period of 21 months, a total of 1001 couples were protected from pregnancy in the project areas due to the program intervention. With regard to contraceptives acceptors, a total of 1936 new and 9922 acceptors were served, and the majority appeared to be pills acceptors (Table 6).

Discussion

Twenty two trained community health agents (CHAs) are the principal distributors of contraceptives in 23 project sites. All the agents except one are males, and have a mean age of about 33 years. About 44, 39 and 17 percent of the interviewed agents had attained primary, junior and secondary schooling, respectively, and all are found to be married. The activities of the CBD agents are supervised by six trained health assistants. One health assistant, on the average, supervises four CBD agents.

The CBD agents, responsible for the motivational and contraceptive distribution activities, motivate the eligibles on family planning at group meetings and through home visits. As to service delivery, the agents distribute contraceptives such as oral pills, condom and foam tablets through the health posts and home visits.

Steering committees at different levels vis, peasant associations, at woreda and regional levels, have been established to enhance community participation for effective implementation of the program. These committees held meetings on quarterly basis. Particularly, the steering committee formed at regional level assesses the overall project performance as well as the budget utilization of the project. It also provides solutions for problems the program encountered during implementation.

This study shows age difference of about eight years between spouses. This difference, although not significant compared to some other African countries, may create communication gap in discussing issues pertaining to family planning and fertility.

One frequently cited barrier to more widespread adoption of family planning in some societies, especially among muslims is the religious opposition (6). Some studies, however, showed that muslims are pro-family planning services. This holds true for the case of Jimma CBD sites where about 97 percent of the sampled respondents are muslims.

Nearly three-fourths of the interviewed women know at least one method of contraception, with the pill as the most frequently (95.1%) mentioned method. The reported level of awareness about contraception is found to be higher by about two percent than the project target of 75%. Generally, a significant increase (about 100%) in the level of awareness of women about contraception is achieved in contrast to the level reported prior to program intervention.

Concerning contraceptive use, proportions of ever and current users are estimated to be 22.7 and 15.9 percent, respectively. These proportions are substantial compared to the reported level (4.4 and 1.3 percent respectively) in the baseline survey (4).

Information on reasons for not using contraceptive methods seem to be important to devise an appropriate IEC and service provision strategies. Desire for more children, followed by disliking of contraception and infertility problems were some of the reasons cited for not using contraceptive methods in the study area. Husbands' opposition and religious reasons, which were mostly mentioned as the main reasons for not practicing contraception in many other areas, have not been considered as hindrances for family planning in the study sites.

In conclusion, the observed high prevalence of knowledge, ever and current use may be attributed to the family planning intervention program that is being implemented at the sites under investigation. If the estimated prevalence rates are accurate, it could be said that the project is successful with regards to providing family planning services compared to the targets set in the

project document. Apparently no major constraints have been observed. Community-based distribution programs can effectively provide services and supplies to communities which otherwise would not have access to family planning services. Therefore, replication of the program in other rural areas should be sought.

For further achievement, it is recommended that:-

- The motivational efforts of the project be continued and strengthened to bring about attitudinal changes among the community towards small family norm with special emphasis to men.
- Films with a variety of family planning messages be made available to the project and be used on a regular basis.
- Since the communities of the study sites are characterized by high fertility, much has to be done to reduce the prevailing high fertility rate by effectively implementing the existing family planning program in the area.
- Consideration should be given to supply health posts with some basic drugs so that the agents would retain a portion from sales of the drugs. This will certainly encourage the CHAs to effectively carry out their duties.
- Sensitization seminars and orientations for influential and religious leaders be organized occasionally, specially at grassroots level, to secure and enhance their support to the program.

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The human trypanosomiasis situation in Gambella, south western Ethiopia

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Abstract: Surveillance of human trypanosomiasis was carried out in Gambella, an endemic region of South Western Ethiopia. The study was conducted in March 1993, October 1993 and April 1994 with the major objective of investigation of the sleeping sickness infection and its vector status in the area. In the present survey no parasitologically confirmed case was detected. The main vectors of the disease, *Glossina pallidipes* and *G. tachinoides* in wooded savanna and forest area and *G. fuscipes* in riverain vegetation, were commonly encountered. Eventhough there were no parasitologically proven cases of sleeping sickness infection due to prolonged combined effect of ecological, climatic and human interference, the presence of potential vectors, *Glossina* species specially along the major river banks, ecological rehabilitation of the area to its previous conditions and the invasion of the game animals might give way to the reappearance of the parasite, *T. b. rhodesiense*. Thus, regular active surveillance of the endemic region is of great importance to control the disease at an early stage before the appearance of epidemics which could be more costly financially as well as in human life. Specific ecological requirements and feeding habits of *Glossina* species, specially of *G. morsitans*, which was not found in this survey, need further investigation. A comprehensive study on community awareness about sleeping sickness and its vector is also recommended to support future control measures. [Ethiop. J. Health Dev. 1997;11(1):23-28]

Introduction

Gambella is the only known endemic area for sleeping sickness in Ethiopia. There was an outbreak of the disease from 1968 to 1970 along the Gilo River where many people died before the disease was recognized as sleeping sickness (1,2). Sporadic cases continued to be detected in the Gilo Clinic and Gambella Hospital (Table 1), after that epidemic.

However, from 1984/85 on wards, Gambella has undergone a drastic ecological change due to extensive deforestation (by settlement, development projects, influx of large numbers of refugees from Southern Sudan) and extensive hunting. These activities have denied the tsetse fly its habitat and thus sleeping sickness cases have decreased significantly.

Since 1991, however, the eco-epidemiological aspect of Gambella is changing to its previous condition. Settlers and almost all the Sudanese refugees, have returned to their original home places. Massive agricultural projects have been terminated or reduced to small scale projects. All these factors would undoubtedly lead to the ecological adjustment of the area to its former conditions which are conducive to sleeping sickness epidemics.

Based on this ecological change of the area, active surveillance was carried out to determine the magnitude of the disease and its vector status. The study was conducted in three consecutive trips,

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March 1993, October 1993 and April 1994 for a duration of one month per trip. The major objective of the study was to investigate the status of sleeping sickness and its vector to help monitor the flare-up of epidemics. Besides, community awareness on sleeping sickness and the vector tsetse fly was assessed.

Table 1: Self reporting cases of sleeping sickness infections from 1976 to 1991 from Gilo Clinic and Gambella Hospital.

Year	No. of cases
1976	38
1977	74
1978	30
1979	15
1980	16
1981	13
1982	11
1983	7
1984	1
1985	9
1986	2
1991	1

Source: Gilo Clinic and Gambella Hospital records

Methods

Study area and population. Gambella is located about 800 Km from Addis Ababa on the Western escarpment of the central Ethiopian massif. It is found on latitude 8°N and longitude 35°E. The elevation ranges from 400 to 600 meters above sea level. The elevation where tsetse fly collection and population screening covered ranged from 480 to 510 meters. The region is divided into six Woredas (=districts), namely Abobo, Akobo, Gambella, Godare, Gokna-Jor, Itang and Jikawo. Four major rivers, (Akobo, Baro, Aluvero, Gilo) and several small tributaries crisscross the region. Of the six woredas only three, (Abobo, Akobo and Gokna-Jor) were known to have tsetse fly infestation and cases of sleeping sickness. The present surveillance activities were conducted at Abobo and Gokna-Jor woredas where several cases of sleeping sickness had been reported since the 1970s. Akobo was not included in this study because it was inaccessible.

According to the 1984 population census the total population of the region was estimated at 65,000 (3), more than 50% of them live in the tsetse fly infested areas. Reports from the Regional Office of Agriculture of Gambella indicated that there were 2500 cattle, 1000 goats in Abobo woreda and 1000 goats in Gokna-Jor woreda. No cattle population are found in Gokna-Jor woreda, a relatively densely forested area with minimum human interference. The indigenous people of the area, the Anuak, live mainly along the Baro, Gilo and Akobo rivers. They cultivate maize and millet at small scale subsistence level. The majority are engaged in hunting, fishing and honey gathering. The young men also go to Damballa and Maji areas in Kaffa for gold mining where sleeping sickness is known to exist.

Climate. The climate is generally warm and humid with a prolonged rainy season which begins in late April and continues until the beginning of November with a mean annual rainfall exceeding 980 mm and relative humidity between 45% and 75%. The rest of the year from late November to the beginning of April is hot, the months of February and March being the hottest. The mean maximum temperature is 36.5°C while the mean minimum temperature is 19.3°C (National Meteorological Service, unpublished report, 1994).

Vegetation. The vegetation of the area is predominantly divided into savannah grassland, wooded savannah and forest land mainly along the Gok area. Vegetation in Abobo is dominated by savannah grassland largely of elephant grass, the *Hyparrhenia* species, while the central part is covered mainly by wooded savannah and partly by forest. Riverain, evergreen vegetation is found along the major river systems and their tributaries. The only protected forest land with big trees such as *Combretum*, *Acacia*, *Ficus* species, etc. is found in Gokna-Jor areas.

Wild Life. Gambella used to be reported rich in fauna, about 39 mammal species were recorded

the majority of which were found in the Gambella National Park which covers 10,800 sq.km. Most of the land protected for game animals as home is not safely guarded. Some of the animals observed during out trips were bush buck, bush pig, wart hog, hyrax, Kling springier and dick dick. Some of them are known as reservoir host for *T.b.rhodesiense*.

Parasitological investigation. Individuals of ages above 10 years from Abobo and Gokna-Jor woredas were selected using random sampling methods and screened for human trypanosomiasis using different parasitological techniques. The methods applied were mainly Microhaematocrit Buffy Coat Technique (MHBCT) (4) as modified by WHO (5), stained blood smears and wet film mounting. The population were all indigenous, the Anuaks. The blood smears stained at the field with Giemsa were examined in the laboratory for the presence of trypanosomes and other haemoparasites at a magnification of 100X.

Entomological methods. During the trips, the area covered for fly collection was about 12,342.72 sq.km and localities included during the survey are indicated in Fig. 1. The first survey was in March 1993 covering villages in Abobo Woreda, 42 km South of Gambella town. The second and third trips were made in November 1993 and April 1994, respectively, covering villages in Gokna-Jor Woreda about 120 km from Gambella.

Biconical traps and moving vehicle were the major methods used in most of the areas where conditions were convenient. Collection of resting and biting flies by hand catch was done in some cases.

Two to five traps were set in every survey site for 24 hours. Attempts were made to set traps in all vegetation types, savannah wood land, forest land and riverain vegetation to collect tsetse flies of different habitats based on their ecological preference. Captured flies were identified using standard keys (5) and subsequently counted and sexed. The collected flies were preserved in 70% alcohol for further study.

Results

A total of 1600 blood samples were collected from different villages and screened (Table 2). No parasitologically proven case of trypanosomiasis was detected. But different species of malaria and blood microfilariae were identified. The known potential vectors of trypanosomiasis, *Glossina* species were collected, identified and sexed as shown in Table 3. *G. fuscipes* was the dominant species along the riverain vegetation; *G. pallidipes* and *G. tachnoides* were collected from thickets of bushes. A decline of the efficient vector *G. morsitans* was observed in the present study.

Table 2: Areas visited and total population screened

Date Surveyed	Woreda	Villages	Total Exam.	Parasite Malaria	Ident. Macrofi.	Remarks
March 1993	Abobo	Ukuna Kiri	140	20	2*	
						MHBCT*
		Ukun kijan	90	10		
		Perbongo	221	24		
		Damessayai	47	3		
		Fukadi	60	8		
		Terkodi	42	4		
October 1993	Goknajor	Tata	115	21		MFBCT*
		Pinybago	39	4		
		Puchala	101	12	1	
		Akima				
		Gilo	35	-		
		Dipa	32	5		
		Pinyudo	178	28	2*	
April 1994	GokanJor	Jinjo	153	27		
		Dipach	65	11		
		Pentini	20	-		
		Gilo area	162	32	1	
		Bottjan	100	13		
		Total	1600	224	6	

*Micro filaria identified by MHBCT

Discussion

It is an established fact that Gambella is the only confirmed sleeping sickness focus in Ethiopia (6,7). However, absence of positive cases during the present survey or low prevalence of the disease reported during the previous studies, does not mean that the disease is eradicated or controlled. This can be attributed to the combination of various ecological, climatic and epidemiological factors that minimized man-tsetse fly-game animals contacts.

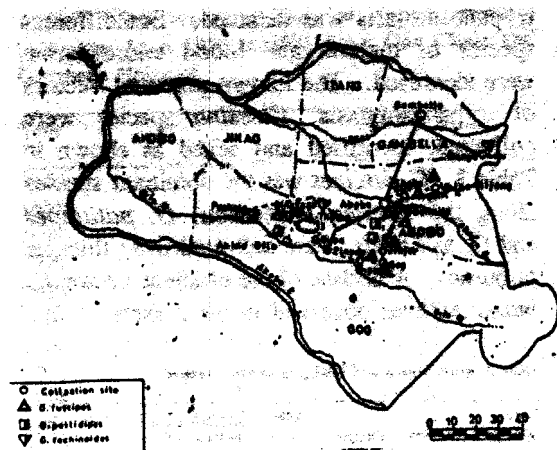


Figure 1: Gambella: villages and tsetse distribution according to the present survey

The epidemiology of human trypanosomiasis is complex. The transmission cycle is subjected to between Man-Glossina-Trypanosome interactions and significantly so in rhodesiense of the sleeping sickness, domestic and game animals involved (8). In our survey, lack of positive isolate has a positive correlation with low density of tsetse flies and game animals. In addition some of the localities previously identified as for sleeping sickness, such as Garri and Akobo, were inaccessible by road.

Table 3: Record of glossina species in Gambella, during three trips.

Species identified	Male	Female	Total
<i>G. Tachinoides</i>	3	1	4
<i>G. Pallidips</i>	11	5	16
<i>G. Fuscipes</i>	192	103	295
Total	206	109	315

It is known that establishments of settlement, state farms, development projects and refugee camps temporarily preclude the tsetse fly from its natural habitat forcing it to retreat to areas where there is less human interference. Extensive changes in the vegetation result from forest clearing, which is an integral part of such projects. The impact of these activities on human trypanosomiasis is significant.

In this survey, we were able to testify that densification of thickets previously cleared for settlement and other development projects had taken place to the extent that the original thicket blocks have now joined together to form continuous ones. This vegetation change has led to the creation of new game habitats and with it new apartment for *Glossina* species. *G. fuscipes* were caught within three kilometers of the state farm along the Aluvero River.

This is a focal point where the laborers take bath and the neighboring villages fetch drinking water. The reservoir host of sleeping sickness, (the bush pig, bush buck, dick dick and the wart hog) were also observed in these localities. Hence, such areas need continuous surveillance and monitoring of the situation regularly. This facilitates the control of peridomestic transmission of sleeping sickness infection by *G. fuscipes* as depicted in other countries (9). In this survey traps were set in all different biotopes where tsetse flies can occupy. However, during the dry season sites around springs, ponds and along the river banks were given more attention, as the flies take shelter in such unfavourable times.

The *Glossina* encountered were *G. tachinoides* and *G. pallidipes* both from open grassland and forest vegetation and *G. fuscipes* dominantly from riverain vegetation. More tsetse flies were collected from riverain vegetation than the savannah and forest types. This might be attributed to the prolonged drought, other climatic and ecological factors in relation to the feeding habit of tsetse flies. Moreover, the disappearance of the efficient vector *G. morsitans* observed during this study needs further investigation.

In a known endemic area, the current strategy for sleeping sickness control is regular active case finding involving population screening and treatment of patients coupled with tsetse fly control (10). Regular surveillance either by a mobile team or trained personnel among the individuals in the area could make it possible to identify the point of transmission and the source of infection. But, in the absence of active surveillance, epidemics may occur at any time. Once an epidemic occurs it will have social and economic consequences which may far outweigh the cost of maintaining surveillance.

Assessment of feasibility of technical, economic and social aspects are all important factors in sleeping sickness and tsetse fly control operation, for which community participation would have a significant impact (10,11).

Investigation of community perception on the relationship of sleeping sickness and its vector tsetse fly, revealed that respondents in age groups below 16 years recognized this fly as a common notorious biting one and identified as "Meio", a vernacular name; but did not have any idea of sleeping sickness. Those in age groups above 16 years, however, knew sleeping sickness, "Okellakai", the local name for the disease. So, the community as a whole could identify the tsetse fly, but did not see it as a vector for sleeping sickness. Respondents in age groups above 30 years, had noticed the seriousness of the disease. Villagers along the Gilo river area had good knowledge about the seriousness of sleeping sickness, because the 1970's epidemic had highly affected the residents. They commented that a lot of tsetse flies were present in the area at that time.

The view forwarded by the majority of the respondents, specially the young age group in the

study, was that sleeping sickness is not a serious disease and should not be given too much importance. This obviously is a wrong impression about the disease. In addition, factors that led to the decline of sleeping sickness in the Region, specially in Abobo and Gokna-Jor woredas, had not been well understood by most of the villagers. That is why they held the view that sleeping sickness is not a serious disease. However, the elders had a notion that during epidemics there had been a lot of tsetse flies in the area and human interference with the ecology (flora and fauna) had been minimum, eventhough they could not understand the role of the flies in the transmission of sleeping sickness. Therefore, there is some background which could be used to explain the vectorial role of tsetse flies and hence introduce methods of control through community participation. We recommend, regular active surveillance of the endemic region to control the disease at an early stage before the appearance of epidemic which is more costly both financially as well as in human life. Secondly, strengthening of treatment centers, introducing effective and sensitive diagnostic methods and trained man- power should be adopted for monitoring the disease in the area. Thirdly, specific ecological requirements and feeding habit of *Glossina* species especially of *G. morsitans* needs further investigations. Inaddition, a comprehensive study on social awareness on sleeping sickness and its vector *Glossina* species is of importance to control the disease.

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Original article

High risk sexual behaviours for STD/HIV, pregnancies and contraception among high school students in a rural town, north western Ethiopia

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Abstract: A cross-sectional study evaluated STD/HIV related sexual behaviours among 383 (90.8% of the required 422) secondary school students in Kolla Diba town of North Gondar Zone using a pre-tested self-reporting questionnaire. Additionally, the contraceptive utilization and occurrence of pregnancies among the 202 female students was evaluated. There were 122 (31.9%) sexually active students, of these 44 were females. Their mean age of sexual commencement was 16.4($\pm$ 2.3 SD) years. The total number of sexual partners in the past six months was around two. Ten (9.3%) had sex with commercial sex workers (CSWs) in the past six months. Only 45.9% had used condoms and half of them regularly. The most common sources of condoms were shops. Fourteen (11.5%) of the sexually active students had contracted a sexually transmitted disease (STD) in the past six months. Three hundred and forty nine (91%) of the students have heard about AIDS, 297 (85.1%) believe in its existence and 166 (47.6%) have seen an AIDS patient. Only 65 (18.6%) felt that they can acquire HIV infection. Of the 44 sexually active female students, 11 (25%) are using contraceptives at present. Oral contraceptive pill is the most commonly used method. The major reasons for the non-utilization of a reliable method of contraception among the non-users were fear of side effects, refusal by the partners and refusal by family. Half of all the sexually active female students were pregnant at least once. Seventy four percent of the female students would prefer to deliver at term if pregnant. Only 37 (18.1%) of the

females favoured the legalization of abortion. A higher level of education was associated with contraceptive use ($p < 0.03$). The implications of these results are discussed. Strengthening the present educational campaign on AIDS, promotion of family life education and making contraceptives accessible in schools are recommended. [*Ethiop. J. Health Dev.* 1997;11(1):29-36]

Introduction

Adolescence is characterized by the period of exploration, experimentation, particularly in relation to sexual activity. Hence, adolescents are exposed to complications of such explorations (1). In Kagera (Nigeria), the sero-prevalence of HIV infection among adolescents (10 -19 years) was 3.2%, of which 63.8% were 10 - 15 years old. In the same study, adolescents who enrolled at school had a higher sero-prevalence compared to non-attenders, 3.0% vs 0.5%, respectively (2). Similar findings were reported from Zimbabwean high school students (3).

In Ethiopia as of April 30, 1995 the cumulative number of AIDS cases reported to the Ministry of Health had reached 18,049. The proportion of AIDS cases among the age groups 5 - 19 years accounted for 5.6% of all AIDS patients (4). Studies addressing sexual behaviours of adolescents in Ethiopia and particularly in Region 3, are non-existent. The few reports on STD/HIV/AIDS related sexual behaviours mainly address the urban population only (5 - 9). Only a handful of

¹From the Department of Community Health, Gondar College of Medical Sciences (GCMS), P.O.Box 196, Gondar, Ethiopia and ² Intern Medical Doctor, GCMS, Gondar, Ethiopia studies on high risk behaviours of rural populations are carried out (10,11). Elsewhere, a report from a study on migrant Ethiopian Jews in Israel showed that the prevalence of HIV infection was 1.74%. Among these, age groups 6 - 17 years contributed 0.1% (12). In a study among 13 - 19 years high school students, over one third had experienced sex mostly with more than one person. The majority of these never used any protective means (5). Hailu (1993) reported that 80% of those high school students who had already started sex were considered at high risk by the investigator's definition (13).

Among school girls, along with STD related problems, pregnancy related complications are also of concern. It is estimated that one million pregnancies occur among adolescents each year in developing countries which represents the apparent high rates of unprotected sexual intercourse, either for pregnancy or STD infection (1). Complications relating to such unwanted pregnancies and induced illegal abortions in African countries are becoming a serious public health problem (14,15).

Despite problems related to unwanted pregnancies, the contraceptive utilization in Ethiopia is still very low (16). A recent study in North Gondar showed that among all adolescent high school students only 5.1% have ever used modern contraceptive methods (17).

Considering problems related to sex and its complications among adolescents, there is an urgent need to document the prevalence of high risk behaviours for STD/HIV acquisition among these teenagers. It is also equally essential to assess the current contraceptive practice and the prevalence of unwanted pregnancies among females. These will assist in identifying areas for intervention. Accordingly, the objectives of this study were to assess the prevalence of problems related to high risk sexual activity, particularly sexually transmitted diseases, and to document the contraceptive utilization pattern among high school adolescent students of a rural town in North Western Ethiopia. It is also aimed at forwarding pertinent recommendations to the concerned authorities.

Methods

This cross-sectional survey was carried out among Kolla Diba high school students in November 1994. All students in Grades 10 - 12 were involved. Grade 9 students were not available on the day of questionnaire administration and therefore, excluded from the study. A pre-tested self-

reporting questionnaire with two major sections was prepared in Amharic and then administered to consenting students. The first section which consisted of 26 questions on basic socio-demographic characters, sexual behaviour, prevailing preventive practices and knowledge and attitude towards AIDS was administered to all male and female high school students. The second, section administered to the female students only, consisted of 28 questions on contraceptive use, knowledge and attitude towards contraception and matters related to pregnancies - their outcome, complications and prevention. All of the students filled out the first section at one time and the same females were requested to fill in the second section on the subsequent day. The overall process was coordinated by 11 senior medical students of Gondar College of Medical Sciences (GCMS).

Prior to distributing the questionnaires, the students were briefed on the purpose of the study and were instructed to provide accurate and honest responses. Confidentiality and anonymity of responses were assured for consenting students.

Data was then compiled and analyzed using a computer EPI INFO version 5 statistical package. Yate's corrected Chi-square test was calculated to compare proportions. Means were compared using the t-test and a One Way Analysis of Variance (ANOVA). Odds ratio (OR) and 95% confidence intervals (CI) were calculated for 2 X 2 tables. Regression and Correlations were run for continuous variables. P values less than 0.05 were considered significant.

Results

Four Hundred and twenty two students from all sections of Grades 10 - 12 responded to the first section of the questionnaire on STD/HIV. Out of these, the responses of 39 students were either grossly incomplete or inconsistent. Therefore, the responses of 383 (90.8%) students were analyzed and reported. For the second section of the questionnaire the responses of 202 female students on pregnancy and contraceptive use are reported.

The socio-demographic characteristics of these students are shown in Table 1. Of all the respondents, 122 (31.9%) were reported to be sexually active, i.e., have had sexual intercourse at least once in the past; out of these 44 (36%) were females. The mean age of sexual commencement was 16.4 ($\pm$ 2.3 SD) (median = 16). The total number of sexual partners per person in the last 6 months were 1.9 ($\pm$ 2.4 SD) (median = 1). Females [mean age of sexual commencement of 15.5 years] become sexually active earlier than boys [16.7 years] (t_1 = 6.7; p = 0.01). Urban students had earlier sex (15.9 years) compared to their rural (17 years) counterparts (t_1 = 32.9; p = 0.01). Males had more sexual partners (mean = 2.2) than females (1.1) (t_1 24.4; p = 0.04). Similarly, urban students had more sexual partners (2.3) than the rural ones (t_1 = 34.5; p = 0.001). Being sexually active was statistically associated with the educational level of the students (X^2 = 15.3; p = 0.0005). Regarding the nature of their partners, 57 (46.7%) claimed to have had intercourse with fellow students, 35 (28.7%) with their spouse, 10 (8.2%) with commercial sex workers (CSW), 2 (1.6%) both with students and CSWs, and 18 (14.8%) did not respond. Within the past one month an average of 2 ± 2.3 (median = 1) episodes of sexual intercourse were reported. Out of the 122 sexually active respondents, 56 (45.9%) had used condoms within the past six months. Twenty two (39.3%) reported to be using condoms always. Seventeen (30.4%) used condoms while having sex with any body, 23 (41.1%) with casual partners, seven (12.5%) with suspicious partners, three (5.4%) with CSWs and six (10.7%) with others. Forty four (78.6%) reported that condoms were readily available. The two most common sources of condoms reported were shops for 25 (44.6%) and the health centre for 19 (33.9%) students. The most common potential problems due to condom use reported were non-approval from the partner by 12 (21.4%) and decreased sexual satisfaction by 11 (19.6%) respondents. The other mentioned problems were unawareness of the appropriate use by four (7.1%), tearing of condoms by three (5.4%), slipping off during intercourse by two (3.6%) and 24 (42.9%) reported none. Of the 122 sexually active students, 14 (11.5%) had contracted STD in the past six months. Gonorrhoea being the most common one in nine, Chancroid in 3, Syphilis and Lymphogranuloma venereum in 2 students.

Local names were used to identify these diseases.

Of the total 383 responding students, 349 (91.1%) have heard about AIDS. More male students knew that AIDS exists than the females [OR = 3.2; 95% CI=1.3,8.0]. Age was thought to confound this result. It was found out that age was associated neither with sex nor with knowledge. Two hundred and ninety seven (85.1%) believed in its existence while 26 (7.4%) did not, and the remaining 26 (7.4%) were indifferent. Younger students (mean = 17.1 years) tended to believe more in the existence of AIDS compared to the older ones (18.3 years) [$F_2 = 17.2$; $p < 0.05$]. One hundred and sixty six students (47.6%) claimed to have seen an AIDS patient and 114 (32.7%) were afraid of getting AIDS. Only 65 (18.6%) felt that they could acquire AIDS. Boys felt more vulnerable to acquire AIDS (OR = 2.5; 95% CI = 1.4, 4.7) and were afraid of getting AIDS (OR = 2.3; 95% CI = 1.4, 3.7) than the girls. Rural students tended to be more afraid of getting AIDS than the urban ones (OR = 1.7; 95% CI = 1.04, 2.8). Finally, those who were sexually active felt more prone to acquire AIDS (OR = 1.9; 95% CI = 1.04, 3.4). The most preferred methods of prevention of acquisition of AIDS mentioned were, having single sexual partner only by 219 (62.9%), using condoms by 64 (18.4%) and abstention from sex by 50 (14.4%) students. Other methods mentioned were having occasional sex, sex with healthy people and with few sexual partners.

Of the same 202 female students who completed the second section of the self-reporting questionnaire on issues related to contraception and pregnancies, 44 (21.8% of all females) were found to be sexually active. Out of these, 15 (34.1%) have used one of the contraceptive methods. The reported methods were oral contraceptive pill (OCP) by 10/15 (66.7%), two had used injectables, one used the rhythm method, others condoms and one did not specify. Out of all the sexually active students, 11 (25%) are currently using at least one of the contraceptive methods. Seven used OCP, two rhythm method, and two used injectable. Sources of these contraceptives were health institutions. Out of the 191 non- users at the moment (these include both sexually active and in-active ones), 45 (23.5%) are planning to use either of the methods in the future. Out of these, 39 were specific about their plans. (Injectable by 16, OCP by 17, the rhythm method by 4, condom by one and Norplant by the other). The three most common reasons for not utilizing contraceptives till to date among the sexually active ever non-users (29) were fear of side effects by seven (24.1%), refusal by the male partner by six (20.7%) and family influence by four (13.8%). Other reasons given were unavailability, cultural influence and religious prohibition. Table 2 shows the known contraceptive methods by the students. Of all analyzed, 33 (16.3%) knew no methods at all. The commonest sources of information about contraceptives were the school for 50 (29.6%) students, mass media for 49 (29%), health institutions for 19 (11.2%) and friends/relatives for 13 (7.7%). The most preferred methods of contraception are also shown in Table 2.

Out of the 156 respondents, 115 (73.7%) would prefer to deliver the baby at term if pregnancy occurred whereas 41 (26.3%) preferred abortion. Twenty seven (65.9%) preferred termination at health institutions. Only 5 preferred induction in the villages and the remaining nine were not sure what to do.

Table 1: **Socio-demographic characteristics of the study population, Kolla Diba, November 1994.**

Variable	Male(n=181)		Female(n=202)		Total	
	Freq.	%	Freq.	%	Freq.	%
Address						
Urban	157	86.8	80	39.6	237	61.9
Rural	24	13.2	122	60.4	146	38.1
Age (years)						
12 - 15	24	13.3	70	34.7	94	24.5
16 - 19	113	62.4	129	63.9	242	63.2
20 - 23	34	18.8	3	1.5	37	9.7
24 - 27	6	3.3	0		6	1.6
28 - 31	4	2.2	0		4	1.0
Marital status						

Single	167	92.3	165	81.7	332	86.7
Married	9	5.0	32	15.8	41	10.7
Divorced	4	2.2	4	2.0	8	2.1
Widowed	1	0.5	1	0.5	2	0.5
Ethnicity						
Amhara	173	95.6	194	96.0	367	95.8
Tigre	4	2.2	6	3.0	10	2.6
Oromo	2	1.1	1	0.5	3	0.8
Kimante	2	1.1	1	0.5	3	0.8
Educational level						
Grade 10	76	42.0	109	54.0	185	48.3
Grade 11	66	36.5	55	27.2	121	31.6
Grade 12	39	21.5	38	18.8	77	20.1

Out of the 44 sexually active girls, 22 reported to have been pregnant at least once. Out of these, 20 had one and two had two pregnancies. Twenty out of the first pregnancies ended up in live births whereas one was aborted and the outcome of the other pregnancy was not mentioned. Both of the second pregnancies were reported to have been aborted. Regarding the possible complications of abortions, 102 (50%) did not mention any. Eighty nine (44.1%) said it could lead to death, sterility by 11 (5.4%) and societal rejection by five (2.5%). Of the 202 students, 37 (18.3%) supported the legalization of abortion whereas 75 (37.1%) objected and 92 (45.5%) were indifferent. Many of those who planned to use contraceptives in the future also supported legalization of abortion (OR = 3.1; 95% CI = 1.28, 7.34). Ever using a contraceptive method was associated with a higher level of education ($X^2_2 = 6.99$; $p < 0.03$).

Table 2: The known (identified) and preferred methods of contraception by the female students, Kolla Diba, November 1994.

Type	Known Methods		Preferred methods	
	Freq.	% ³	Freq.	% ³
None	33	16.3	100	49.5
OCP	146	72.3	81	40.1
Rhythm	121	59.9	54	26.7
Condom	87	43.1	43	21.3
Breast feeding	38	18.8	22	10.9
IUD ¹	29	14.4	15	7.4
Abortion ²	9	4.5	-	-

¹ Intra-uterine device

² Abortion was mentioned as a means of contraception.

³ (N = 202)

Discussion

This Study has attempted to assess the prevalence of high risk and protective sexual behaviours, contraceptive utilization and pregnancy related matters of high school adolescents in the rural town of Kolla Diba, Dembia District of North Gondar, Ethiopia.

The basic demographic characteristics of the study population are not different from other similar school populations (5,17,18), except that the overall proportion of urban students is very high compared to the national population proportion.

Though the majority of the sexually active students had one partner, the mean was found to be around 2 partners with large variance. This indicates that the rate of promiscuity is high among these adolescents. This can predispose them to acquisition of STDs/HIV (19,20). The high number of sexual partners of adolescents may be the result of the great perceived value of an exciting life through sexual pleasure (21). In this study, the presence of high risk factors for acquisition of STDs (22,23) among the adolescents is evident, these being multiple sexual partners, sex with CSWs and failing to have a protected sex. Though the proportion of sexually active female students was comparable with other parts of Ethiopia (5,17,18), females in this study demonstrated earlier sexual commencement compared to the males, even compared to other parts

of the world (24). This early commencement may lead to high rates of unintended pregnancies and their complications.

Around half of the sexually active students reported using condoms and again only half of these used them regularly. This figure seems higher compared to earlier reports (8,10,11), yet, we feel that the utilization of condoms is still not satisfactory. It requires intense effort to convince these group of adolescents to adopt the use of condoms. The fact that the majority of condom users obtained them from shops can be viewed from two opposite directions. On the positive side, it seems a good indicator of familiarization of condoms among the population and making it easier to obtain them from shops on streets rather than going to health institutions. On the other hand, if condoms are not properly stored in these shops before getting dispensed, then their potency should be questioned.

A substantial proportion of the students have heard about AIDS and believe in its existence which is obviously higher compared to findings from a study among nearby rural farmers (11). This is an encouraging finding which may be due to the presence of AIDS Club in the school as well as the health education sessions being conducted by various health personnel. It is interesting to find out that almost half of all the students claimed to have seen an AIDS patient. This may show how much people have become aware and sensitive in identifying those having the disease. Probably it may have also contributed to strengthening the belief in the existence of the illness. On the other hand it is discouraging that less than a quarter had the fear and perception of getting AIDS. Our finding was even more discouraging compared to those of other studies (10,11). This can have a very negative influence on reducing high risk behaviours, thus, becoming a major risk factor for HIV acquisition (1,23).

Ever use of contraceptives by female students (34.1%) in this study, seems low compared to the previous report from Gondar (17) and Kenya (25), yet comparable with that of the general population of Gondar (26) but much higher than students in Harar in 1992 (18), Jimma (27) and Banjul, the Gambia (28).

It is worrisome to note that only 25% of the sexually active females were using any method of contraception. Moreover, very few (23.3%) of the currently non-users are planning to use them. The most commonly attributed reasons for non-utilization of contraceptives among such adolescents (in our study as well as others) are fear of side effects, refusal of the partner, family influence, lack of knowledge, being shy to use, no desire to use, inaccessibility and desire for more children (17,18,26,29). Hence, in all family life education programs it is vital to incorporate these issues and minimize the misconceptions, which obviously can lead to higher rate of utilization by the clients.

In this study 50% of the sexually active students claimed to have been pregnant. This figure seems very high and reportedly exaggerated compared to other reports from Gondar (17) and Harar (18). This is alarming because of the multiplicity of problems related to school-girl pregnancies. Students usually forget that unprotected sex can lead to pregnancy. Moreover, they have insufficient knowledge about the use of contraceptives and they have conflicting messages on the reproductive role of young women and their low self-esteem in their interaction with older, experienced male partners (30).

It may be interesting for policy makers that only 18.1% of our study population and 5.1% of the female population of North Gondar (26) favoured the legalization of abortion in Ethiopia. This may be mainly due to the strong religious and traditional background of the population in this region.

Level of education was associated with contraceptive use in this study, which was also observed else-where (25,26,28,31-34). Therefore, the role of formal education in the promotion of contraceptive use and family planning among adolescents should be beyond doubt.

In general, though the majority of the students knew about AIDS, the perception of risk of acquisition is weak and the practice of protected sex is also low. Contraceptive utilization among

adolescent school girls is also low and the rate of pregnancies is very high. Therefore, the present effort of educating students on AIDS and other STDs needs to be intensified so that it can bring about favourable attitudinal change and subsequent behaviour modification. Moreover, promoting family life education in schools can be very useful. Contraceptive technology should be made more readily available and accessible to the needy school population. These have also been consistently recommended by several other authors (18,24-26,35,36), hence, deserve the due attention of the responsible policy makers and implementers.

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The nominal group technique for participating communities in analyzing rural town water and sanitation situation

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Abstract: To explore sanitation problems and their causes, a series of independent group discussions was conducted with residents of a rural town stratified into seven social levels using the Nominal Group [Discussion] Technique (NGT).

Indiscriminate open field defecation and garbage disposal were the two most mentioned and ranked problem items followed by unsanitary food and drink services. The top identified cause items were absence of public latrines, failure of the municipality to control town sanitation and absence of solid waste disposal facilities.

A Combined group gave the highest Median Agreement Score (MAS) of 10. The smallest median agreement was scored by Youth and Health workers' groups (MAS=6.5) for mentioned, and by youth and Ordinary residents' groups (MAS=4.5) for ranked problem items. Health workers' group (group 7) ranked MAS of 5.

The MAS for causes of sanitation problems of the Combined group was 8 for mentioned, and MAS of 4 for ranked items.

Development workers ought to give due recognition to communal stratification when making need assessment for better realignment of diverse view points and interests during project development. Modifying and validating NGT for a broader use in assessing community health problems and needs is suggested. [*Ethiop. J. Health Dev.* 1997;11(1):37-42]

Introduction

Poor environmental sanitation and lack of safe and adequate water supply are known to adversely affect human health(1,2,3). At present, the sanitation and water supply situation in most Ethiopian towns, including the capital Addis Ababa, is very poor(4).

To improve this situation, the search for a practical and acceptable method of community participation is vital(4,5,6). The first essential step in community participation is intensive consultation with the community(7).

Different approaches of community participation and consultation have been used in environmental health activities(8). These include seeking contribution, forming self help groups, deploying community specialized workers and mass action. Consultations have been done by simply talking to local officials, influential people, interviewing all or a segment of the resident population or by calling public meetings.

Communities in both small and large towns are composed of families and groups of diverse Psycho- socio-cultural, demographic, economic and political backgrounds(8,9). This stratification can represent a more or less distinct spheres of interest which merge as one on some and conflict on yet other communal issues.

Despite underdevelopment of health services management and incorrect community participation

approaches, it has become a tradition to attribute failure of projects to unpreparedness (unwillingness) of communities to participate.

The Nominal Group Discussion Technique (10) has the advantage of bringing different people to a designated discussion forum where each participant silently generates ideas and forward them to the floor with equal opportunity and pursue them without being threatened from (but realigning himself with) other discussants; as such assessing real feelings and objective experiences. This is unlike other assessment methods, such as focus group, formative evaluation and rapid methods, where opinions of dominant (influential) individuals may be more expressed and/or ideas may be heavily influenced by that of investigators and data collectors.

In this study, the NGT is used to identify sanitation problems and their causes as perceived by residents across societal groupings in order to initiate action to improve the town's sanitation; also the application of the NGT as a method of exploration into communities health problems in urban settings is appraised.

Methods

The study was conducted in a small town of Koladiba north west Ethiopia with a population of some 6000. The first national health center was opened in the town owing to a large number of deaths from a malaria epidemic in the Dembia plain. Following that, for over 40 years now, the area has served as field training site for different categories of health science students with their massive inputs of health programmes, including in water and sanitation. Residents who lived in the town for at least one year, who are older than 15 years of age and are able to read and write were the source population for the study. For the purpose of sample selection for the study, after sufficient consultation with informants, seven different stratum (interest groups) were identified. These were:

- Group 1. Women group
- Group 2. All development agencies
- Group 3. Youth group
- Group 4. Town administration
(authorities)
- Group 5. Influential people
- Group 6. Ordinary residents
- Group 7. health workers

Additionally,

- Group 8. Combined group and
- Group 9. Expert group were formed.

After dividing the town into seven geographic zones, using informants, one participating influential, youth, woman and ordinary resident were identified by walking door to door and invited for the group discussion from each zone. Health workers group was composed by representation of all professional categories from the health center and a community health agent in the town. Town authority group was made up of individuals from the different sections of the municipality, district administration, court and police offices. Development agencies group was a representation from district education, agriculture, natural resources, finance offices, schools and Amhara Development Association.

Combined group was formed by selecting one person from each of the seven groups for repeat discussion on each issue. Expert group was made up of knowledgeable (about the town) academic staff of the different departments of the Gondar College of Medical Sciences and Kola Diba health

center. The number of discussants in each group varied from six to eight on different days.

Data Collection Procedure: After a brief introduction on the purpose and conduct of the study to a gathering of all participants by the principal investigator, each of the seven nominal groups was led to its respective discussion room. A trained medical doctor or nurse was assigned for each group to lead and moderate the discussion according to a modified nominal group discussion procedure (Appendix 1). For four mornings in a row the seven groups discussed the four identified water and sanitation issues (Appendix 2) simultaneously in different rooms. Individuals selected from the seven groups formed a Combined group and discussed the same issue on four afternoons. There was no payment for participation except providing tea and cookies during breaks.

Data Analysis: Ideas listed down by each group at the end of step 3 were reorganized into categories of sanitation problem and cause items by the investigators such that all ideas stated by the nine groups are accommodated. The categories thus formed were used as data for analysis. If a category is included in the list of ideas made by a group, we called it MENTIONED ITEM. Out of the list of mentioned items, the ones selected as 10 most important by a group on step 4 were named RANKED ITEMS.

Results

All together the ideas brought by the nine groups could be organized into 20 sanitation problems and 22 cause/reason items.

Tables 1 and 2 show the top five mentioned and ranked sanitation problem and cause items identified by all groups, respectively.

Uncontrolled (open field) human excrement and garbage disposal were the two problems mentioned and ranked by all the nine groups. Unsanitary food and drink services have also been mentioned as problems by all but ranked by seven groups.

The two items at the top of the list of causes of sanitation problems of the town were: absence of private and public latrines and lack of sanitation control by the municipality for mentioned, and lack of sanitation control by the municipality and absence of solid waste disposal facilities for ranked, items.

Items that received least attention (mentioned or ranked by two or less groups) were: absence of drainage system and poor housing condition as problem items, and misuse of collected sanitation revenue(tax) by the municipality, overall underdevelopment and poverty of residents, lack of coordination between concerned agencies, and poor sanitation of the prison located in the centre of the town, as cause items.

13(65%) and 8(42%) of identified sanitation problems were similarly mentioned and ranked respectively, by five or more groups.

Out of 36 possible pairs (combinations) that could be formed among the 9 discussion groups for similarity analysis, groups 4 and 5 mentioned the highest (11) and groups 4 and 6 ranked the highest (10) similar problem items. Likewise, groups 4 and 7 mentioned the highest (12) and groups 4 and 5 ranked the highest (7) similar cause items.

Least similar pairs (similar in four or less items) were groups 1 and 3 for mentioned and 1 and 3, 2 and 6, and 3 and 6 for ranked problem items, and groups 2 and 5 for mentioned and 1 and 6 for ranked cause items.

The Combined discussion group had the highest MAS of 10, equal to group 4, for mentioned and highest score of 6 equal to group 1, for ranked problem items. The smallest MAS was scored by groups 3 and 7 (6.5) for mentioned, and by groups 3 and 6 (4.5) for ranked problem items. While the highest MAS for ranked items was seven, Health Workers group ranked 5th.

Table 1: Top 5 mentioned and ranked sanitation problem items of Kola Diba town

Rank	Mentioned Item	Mentioned by,	Rank	Ranked Item	Ranked by,
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		# of groups			# of groups
1	Uncontrolled open field defecation	9	1.	Uncontrolled Open Field Defecation	9
1.	Uncontrolled Garbage Disposal	9	1.	Uncontrolled Garbage Disposal	9
3.	Absence Of Private & Service Latrines	8	1.	Absence Of Private & Service Latrines	9
4.	Unsanitary Food & Drink Services	7	1.	Unsanitary Food & Drink Services	9
5.	Uncontrolled Disposal Of Dead Animals	6	5.	Absence Of Abattoir	8
5.	Absence Of Abattoir	6			
5.	Absence Of Public Latrines	6			

The MAS for cause of sanitation problem of the combined group was 8 equal to groups 7 and 9 for mentioned, and 4 equal to groups 4, 5 and 9 but higher than all other groups, for ranked items. Group 5 has the lowest MAS (5) for mentioned, and groups 2, 3 and 7 the lowest (3) for ranked, cause items.

On a composite scale of the top five list of -

1. mentioned items of the combined group
 2. mentioned items of all groups, and
 3. ranked items of all groups,
- uncontrolled open field defecation, disposal of dead animals and garbage appeared as common sanitation problems, and lack of attention from the municipality, absence of solid waste disposal facility and unimplemented town's master plan as common cause/reason items.

Discussion

While four of the 20 sanitary problems are of related educational and administrative nature, 16 are considered strict sanitation problems. Participants have agreed that the town exhibits a lot of deviation from basic sanitary standards.

The composition and scope of the listed items, contrasted to that of the experts, gives a good coverage and assessment of the towns sanitation problems and their causes.

Before reorganizing the mentioned ideas into item categories by the investigators, we have been able to see that the participants have spoken out their subjective and objective experiences on the discussion issues in their own special way. Not unsurprisingly, the observation which motivated and compelled the investigators to undertake this study, uncontrolled massive open field defecation and garbage disposal practice, has also been identified as the most important sanitation problem by the discussion groups. Similarly, absence of human waste disposal facilities (private and public latrines) has been the top identified cause for the poor sanitation in the town. By this, one can be assured that, at least, there is no perception gap (ignorance), on the side of the participants, in accepting the current sanitation status of the town as poor.

Discussion participants have shown to be particularly unhappy (distressed) about the poor sanitation of food and drink services. The realization that they are public services compounded by the fear that they may serve as a common source of infection might have heightened the concern. Also in this connection, absence of slaughter house (abattoir) resulting in unsanitary out door animal slaughtering for public meat supply in poorly cared for butcher houses is noted as an important (top 5) mentioned and ranked item.

Table 2: **Top 5 mentioned and ranked cause items for the sanitary problems of Kola Diba town**

Rank	Ranked item	Ranked by, # of groups	Rank	Mentioned Item(#)	Mentioned by, # of groups
1.	Lack of Attention From Municipality	8	1.	Absence of Private & Public Latrines	8
2.	Absence of Solid Waste Disposal	6	1.	No Control On Food & Drink Services	8
3.	No Control On Food & Drink Services	5	3	Absence Of Solid Waste Disposal	7
3.	Absence of Abattoir	5	3.	Unimplemented Town Master Plan	7
3.	Unimplemented Town Master Plan	5	3.	Absence Of Abattoir	7
3.	Failure in Giving & Receiving Health Educ.	7			
3.	Lack Of Attention From Municipality	7			

When discussing the cause, participants primarily pointed to the failure of the municipality to discharge its coordinating and controlling responsibility in sanitation. This message speaks against the often heard controversy over whether the municipality, the health centre (MOH), or the public is to be charged (blamed) for sanitation of rural towns. The participants have also defended themselves against the inadvertent criticism and conclusion by administrators and health workers that cause of failure of health projects is lack of community participation and/or awareness.

The fact that a good number (usually >60%) of items are similarly mentioned by five or more groups shows that participants fairly share the way they perceive and realize the town's sanitation problems and their causes.

It is interesting to note that group 8 (combined) had the highest MAS for both mentioned and ranked sanitation problems and cause items. This means, the repeated and combined nominal group discussion has well accommodated and realigned ideas from different backgrounds. Indicating the need for assuring the balanced representation of important strata in society in development consultation exercise.

The consistently highest pair similarity and high MAS reached by group 4 (town administration) shows that they fairly share and understand residents' opinion on sanitation problems and their causes. An attribute that could have emanated from their ability to list more problem and cause items than the other groups, perhaps as a result of frequent contacts they had working in the municipality and other local government offices.

It is alarming that health workers group (7) repeatedly scored in the least category of MAS for both sanitation problem and cause items; suggesting a rather low degree of understanding and sharing communities' feelings and experiences on sanitation issues.

The fact that 3 out of 5 - 6 top sanitary problem and cause items (mentioned and ranked) were commonly picked as priority items by the 3 independent scales shows the seriousness of the demand to improve the towns sanitation in this selected areas. Further more, the consistency of the items to appear in three different measurements qualifies the stratified nominal discussion procedure as a reliable explorative method for use in community health (sanitation) diagnosis.

The major limitation of this technique is that it involved people who are able to read and write only. Hence, it is recommended that the method be modified so as to make it simpler and workable with illiterate people too. This is especially necessary during the silent generation and presentation of ideas in this technique.

The town's administration and health authorities can use the list of items produced in this study to plan improved sanitary services for the town. The municipality should take the primary responsibility in managing sanitation activities of the town. To exercise control of sanitation in the town the municipality might have to devise a mechanism to ensure observance of at least minimal sanitary standards. Front line health workers should be able to establish good working relationship and acquire skill on community need assessment to be able to reflect on real feelings of their catchment population.

Sanitation project initiators should consider societal differentiation to ensure their fair representation beginning at the stage of situation analysis. And finally, NGT needs to be further

validated for use in quick and reliable exploration of public interest in water and sanitation and other community health programmes.

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Our gratitude also goes to residents of Kola Diba town and staff of Kola Diba health centre who willingly and unfailingly took part in a series of often tiresome nominal group discussions.

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Appendix 1. procedure of the nominal group discussion technique

The research assistant distributes the Nominal Group Task Statement Form, reads out the discussion issue on it and advises the participants to follow him for the NG steps.

STEP 1. *Silent Generation of Ideas*: each participant silently generates ideas and writes them down on the NG task statement form.

STEP 2 *'Round Robin' Feedback of Ideas*: the research assistant asks each participant in turn to contribute one of the ideas he/she has written until each finishes his ideas. Each idea is numbered and written on a flip chart.

STEP 3. *Serial Discussion of Ideas*: each idea listed will be discussed in turn. The discussion's objective being to obtain clarity and to air out points of view, but not to resolve differences of opinion.

STEP 4. *Final Vote*: each participant selects 10 ideas which he thinks are most important from the total and list them on to a separate sheet of paper and rank them from one to the highest(usually ten).

Appendix 2. nominal group discussion issues

DAY 1. a. What water problems do you see in the town?

b. What sanitation problems do you see in the town?

- DAY 2. a. What are the causes/reasons for the town's water problems?
b. What are the causes/reasons for the town's sanitation problems?
- DAY 3. a. What should the town's administration do about water and sanitation?
b. What should residents of the town do about water and sanitation?
- DAY 4. a. What should the health centre do about water and sanitation in the town?
b. What should other health agencies(like the GCMS) do about water and sanitation in the town?

Pneumonia and malaria in under-five children of southern Ethiopia

Solomon Tesfaye¹, Derege Kebede² and Sally Stansfield²

Abstract: An observational and experimental study was conducted to see the proportion of overlap between the clinical and laboratory diagnosis of pneumonia and malaria and the therapeutic effectiveness of cotrimoxazole. Children under five years who presented to the clinic with fever and/or cough or difficulty in breathing were enrolled in the study. Blood smears were done for all enrolled children while chest x-rays were obtained only for those children with parasitaemia or who met the clinical case definition for pneumonia. There were 736 children who met the clinical case definition of malaria, while 731 met the clinical case definition of pneumonia. Of these two groups, 456 (61.9%) of children with clinical malaria and 62.3% of those with clinical pneumonia represent the overlap of children who met both clinical definitions. Specific clinical definition did not differentiate pneumonia and malaria. A single treatment of both diseases by cotrimoxazole is showed to be as effective as a combination of treatment with chloroquine and procaine penicillin. Parasite clearance was better in the cotrimoxazole group than the chloroquine-penicillin group, however the cotrimoxazole group had a higher rate of recurrences with two out of 41 patients smear positive at day 14. A significant proportion of overlap in the clinical and laboratory diagnosis of pneumonia and malaria is shown and that a three days cotrimoxazole therapy is equally effective in treating both diseases as a combination of chloroquine and procaine penicillin is. [*Ethiop. J. Health Dev.* 1997;11(1):43-50]

Introduction

Pneumonia and malaria are the major causes of mortality and morbidity among children in developing countries including Ethiopia (1-5).

Malaria treatment is recommended for a child living in a malaria endemic area, where microscopy is not available if the child has fever or history of fever with no other obvious cause. In highly malarious areas, malaria treatment is advised for all children with fever whether or not other obvious causes are present (6). Treatment for bacterial pneumonia is recommended for any child with cough or difficulty in breathing who has a high respiratory rate or lower chest wall indrawing(7).

Studies conducted in several countries indicate that there is a significant overlap in the clinical presentation of pneumonia and malaria (8-11). Case management algorithms are based on the premise that a child is likely to have only one disease at a time. Thus a child with fever and cough may meet clinical case definition for both malaria and pneumonia but have only one disease, or none at all and yet receive one or two treatments unnecessarily. Conversely a child may have both disease but be assessed and treated for only one.

Cotrimoxazole, which has a similar effect of pyrimethamine/sulfadoxine (fansidar), is known to be an effective antimalarial in adults and children over 5 years of age. Because of its broad

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spectrum efficacy, low cost, ease of administration and low incidence of adverse effects,

cotrimoxazole is the preferred drug in most settings for out patient treatment of bacterial pneumonia. A study of subjects with *falciparum* parasitaemia in Tanzania (12), Mozambique (13) and vivax malaria (14), have reported cotrimoxazole as being effective as chloroquine.

Better definition of the patterns in the clinical presentation of these diseases is needed to permit more effective diagnosis and treatment. Documentation that a single drug is effective against the pathogens of both diseases could both save money and avert deaths.

This paper is a report of a study which was designed to define the overlap in the clinical presentation of pneumonia and malaria and to determine the response to cotrimoxazole treatment of children with these diseases.

Methods

An observational and experimental study (a randomized, double-blind clinical trial) was conducted between September to December 1993 when rates of pneumonia and malaria reach a maximum, according to reviews of health institutions records.

The study was conducted in the two health centres and one hospital located in Awassa Town. Awassa Zuria district has an area of 1875 sq.km with a total population of 307,767. Approximately 23% of the population is urban, and children under five years of age account for 19% of the total. The majority of the population are farmers, and 75% live at altitudes ranging from 500-3500 m. Awassa lake is found in the northwest central part of the district with an area of 129 sq.km and average depth of 10 metres. Most of the area are *Kola* with annual temperature ranging from 4.5 to 34 degree centigrade. There are two rainy seasons in the area, one with heavy rainfall in the month of July to September and the other with light rainfall from January through March. The annual rainfall averaged 1110 mm.

A nurse asked each mother or other care taker why the child had been brought to the clinic, and up to three reasons were recorded for each child. Every child brought to the clinic because of fever and/or cough or difficulty in breathing whose mother gave verbal informed consent for participation, was enrolled in the study. Children with severe cases of pneumonia and malaria or those who had any history of taking antimalarial medication in the past two weeks were considered ineligible for the clinical trial.

At enrolment, care-takers were interviewed to obtain data including the name, address, age, sex, history of fever, cough, difficulty in breathing, rapid breathing and any medication taken, physical examination and laboratory tests.

The axillary temperature was taken using an ordinary mercury thermometer for at least one minute and respiratory rate was counted for one full minute when the child was calm or undisturbed. Weight and height were measured using a standard scale in the health institutions. Using the clinical history and physical findings, the children were categorized into groups based on whether they met the clinical definition of malaria and/or pneumonia.

After the clinical diagnosis was recorded, the children under-went laboratory investigation. Haemoglobin in gm/100dl was determined for all children using Sahel's method. Thick and thin blood films of all enrolled children were made and stained with Giemsa, then examined under oil immersion. Asexual forms of *plasmodium* were typed and their density assessed by counting against 200 white blood cells. Densities were expressed as parasites/mm³ assuming 8000 WBC/mm³ of blood. Blood films were reported as negative only after examination of 100 high power fields. Chest radiography was performed only for children with parasitaemia or for those meeting the clinical definition of pneumonia. Children who didn't have malaria parasitaemia and who didn't have radiography were assumed to have negative chest radiographs. Laboratory technicians and radiologists were not aware of the clinical diagnosis and were not allowed to see any child before completing diagnostic tests.

Based on results of the laboratory and chest x-ray investigation, patients were given final diagnosis. Mothers or other caretakers of children with both laboratory diagnosed malaria and radiographically diagnosed pneumonia were invited to be participants in the clinical trial.

Participating children were randomly allocated into the two treatment groups using table of random numbers.

Group one received cotrimoxazole at standard dosage (trimethoprim 6mg/sulphamethoxazole 30mg/kg body weight in two divided doses daily, administered orally as syrup, crushed powder or tablets) for three days.

Group two received the standard regimen of chloroquine base (10mg chloroquine phosphate base/kg of body weight followed by 5mg/kg 6-8 hours later and 5mg/kg on each of the second and third days orally as syrup, crushed powder or tablets) and procaine penicillin at a dose of 50,000 units/kg body weight intramuscular daily for five days.

All patients were invited for a return visit on the 7th and 14th days after initiation of treatment. To improve compliance, the cost of medication, investigation and transportation were covered by the research fund and it was explained that the child would be seen at each visit with the minimum of waiting time.

During each follow up visit, parents were asked about the compliance with the medication regimen and whether their child had been ill or had experienced fever, cough, difficulty in breathing, vomiting, diarrhoea or poor appetite. The child was examined for the presence of chest indrawing and other danger signs. The temperature and respiratory rates were recorded and blood film performed.

Malaria therapy was considered to have been successful if no parasites were detected on day seven and 14. All patients with parasitaemia on day seven were treated with a single dose of Fansidar (25mg pyrimethamine and 500 mg sulfadoxine).

A child was considered ill if he or she had fever and/or one more symptom of vomiting, diarrhoea, cough or loss of appetite or three of these symptoms without fever. The overall condition was recorded as worsened, same or improved.

Clinical malaria was diagnosed in the presence of a fever or history of fever. Fever was defined as an axillary temperature of 37.5 or more. Clinical pneumonia was diagnosed in the presence of chest indrawing, cough or difficulty in breathing with a rapid breathing rate of 60 per minute or more for children less than two months of age; 50 or more breathe for children 2-11 month of age; and 40 or more for children 12 months of age or older. Severe pneumonia was diagnosed if chest indrawing was present.

Table 1: **General characteristics of the study population, Awassa (southern Ethiopia) 1993.**

Characteristics	Number (%)
1. Sex	
male	601 (54.5)
Female	492 (45.5)
2. Age (months)	
0-12	439 (40.2)
25 - 36	212 (19.4)
37 - 48	158 (14.4)
49 - 60	152 (14.4)
3. History of medication	
Antimalarial	243 (22.2)
Antibiotic	13 (1.2)
Procaine penicillin	98 (9.0)
Chloramphenicol	41 (3.8)
Bacterium	1 (0.1)
Ampicillin	4 (0.4)
Other	51 (4.7)
Others	1 (0.1)
4. Smear positive	253 (23.1)
Type of <i>plasmodium</i>	
<i>Falciparum</i>	88 (34.8)
<i>Vivax</i>	165 (65.2)
Parasite density	
1 - 4999	87 (34.4)
5000 - 9999	99 (39.1)
10000 - 14999	29 (11.5)
15000 - 19999	12 (4.74)
≥ 20000	26 (10.5)
5. Nutritional status	
Stunted	282 (25.8)
Under weight	230 (21.0)
Wasted	228 (20.9)

Laboratory diagnosis of malaria was made in presence of asexual parasites in the blood smear. Cases of severe malaria included those children with parasite density exceeding 250,000/ mm³ and with a haemoglobin level of 7.5gm/100ml or less. Radiographic pneumonia was defined as the presence of parenchymal density compatible with pneumonia as interpreted by the radiologist. Radiographs showing lobar consolidation or involvement of more than one lobe were classified as severe.

The Epi Info (version 5) and the Statistical Analysis System (SAS, version 6.04) programs were used to analyse the data collected. Data on the parasite density were transformed to natural logarithm scale before they were used in statistical analysis.

Parents/care takers of the potential children for the study were informed about the aims, methods, anticipated benefits and potential hazards of the study and verbal consent was obtained. Severe cases of either pneumonia or malaria were admitted and treated appropriately and considered ineligible for the clinical trial. The study was approved by both a review committee of the Department of Community Health and the Research and Publications Committee of the Faculty of Medicine.

Results

A total of 1093 children were enrolled in the study. A summary of the general characteristics of the study population at the time of entry into the study is summarized in Table 1. The average age of children enrolled was 25 months.

Of the total children enrolled, fever, cough or difficulty in breathing or both were reported in 305 (27.9%), 218 (19.9%) and 570 (52.2%) of the children by mothers or other care takers, respectively. The parasite densities ranged from 130 to 320000, but only three cases had a parasite density of more than 100,000. Among those who had history of taking procaine penicillin and

ampicillin, 22% and 7.8%, respectively, and also 14.8% of those taking other medication were found positive for plasmodium. Of 13 who had history of taking antimalarial two (15.4%) had positive smears, of which one was *falciparum* and the other *vivax* malaria.

Of the total of 843 children who had radiographic investigation, 403 (47.8%) had positive findings. Among these, 33 (8.2%) were severe pneumonia, 360 (89.3%) none severe, and 10 (2.5%) had other diagnosis. Among those who had history of taking antibiotic, 38.8% were found to have radiographic pneumonia, while 15.4% of those who had taken antimalarial had radiographic pneumonia. Six children who had history of taking injections of procaine penicillin were found to have both radiographic pneumonia and parasitaemia.

There was significant overlap in clinical as well as laboratory diagnosis of pneumonia and malaria. The clinical definition of both pneumonia and malaria was met by 456 (41.7%) children (Table 2).

Table 2: Number of children under five with fever and/or cough or difficulty in breathing meeting clinical and laboratory definition for malaria and pneumonia, Awassa (southern Ethiopia) 1993.

Clinical	Number (%)	Laboratory	Number (%)
1.Malaria alone	280 (25.6)	a. Malaria	81 (28.9)
		b. Both	14 (5.0)
		c. Neither	185 (66.1)
		Total	280 (100)
2.Pneumonia alone	275 (25.2)	a. Malaria	11 (4.0)
		b. Pneumonia	107 (38.9)
		c. Both	15 (5.5)
		d. Neither	142 (51.6)
		Total	275 (100)
3.Both	456 (41.7)	a. Malaria	50 (10.9)
		b. Pneumonia	176 (38.6)
		c. Both	81 (17.8)
		d. Neither	149 (32.7)
		Total	456 (100)
4.Neither	82 (7.5)	Neither	82
Total	1093(100)		

Children who satisfied the clinical definition for both pneumonia and malaria were more likely to have radiographic pneumonia (OR=4.76, 95%CI: 3.62-6.26), parasitaemia (OR=1.75, 95%CI: 1.31-2.35) and both at the same time (OR=4.53, 95%CI: 2.85-7.23) than children who do not meet the definition.

A total of 107 out of 283 (37.8%) with radiological signs consistent with pneumonia and a negative blood film, and 15 out of 110 (13.0%) with radiological signs consistent with pneumonia and a positive blood film, satisfied the clinical case definition for pneumonia.

However 61 out of 142 (43.0%) of those satisfying the clinical case definition of malaria with a positive blood film and no radiological evidence of pneumonia, also satisfied the clinical case definition of pneumonia.

The likelihood of radiographic pneumonia was higher in smear positives than smear negatives (OR=1.53, 95%CI: 1.15-2.04). There was no difference observed in relation to age, sex, haemoglobin, nutritional status, type of parasite or parasite density.

The comparability of the children in the two treatment groups is demonstrated in Table 3. There was no significant difference in their general characteristics except for weight-for-height z-scores. Only three children were lost in the follow up.

Table 3: General characteristics of the treatment groups, Awassa (southern Ethiopia) 1993.

	GROUP-1	GROUP-2	P-VALUE
Number of children	42	52	
1. Age mean $\pm$ sd	28 $\pm$ 17.6	31 $\pm$ 17.7	p=0.60
2. Sex Male	20	29	
Female	22	43	p=0.43
3. Temperature Febrile	35	44	

Afebrile	7	8	p=0.87
4. Haemoglobin			
mean $\pm$ sd	8.68 $\pm$ 1.27	8.7 $\pm$ 0.88	p=0.91
5. Nutritional status			
mean $\pm$ sd HAZ	-0.31 $\pm$ 1.92	-0.59 $\pm$ 2.06	P=0.50
WAZ	-1.42 $\pm$ 1.40	-1.03 $\pm$ 1.29	P=0.17
WHZ	-1.41 $\pm$ 1.13	-0.65 $\pm$ 1.69	P=0.01
6. Density			
log.mean $\pm$ sd	3.823 $\pm$ 0.369	3.914 $\pm$ 0.481	p=0.30
7. Type			
<i>Falciparum</i>	16	23	
<i>Vivax</i>	26	29	p=0.55

Treatment in group-1 = Cotrimoxazole; group-2 = Chloroquine + Procaine

WHZ= Weight-for-height z-scores; WAZ= Weight-for-age z-scores; WHZ= Height-for-age z-scores.

During the first scheduled follow up visit on the 7th day of treatment, those taking cotrimoxazole alone had a higher risk of fever and fast breathing but had less risk of having cough, vomiting, diarrhoea or smear positivity than those taking procaine penicillin and chloroquine. The proportion of children who clinically improved was higher in group-1 than group-2, although the difference was not statically significant (Table 4).

At the time of the second follow up visit on day 14, those treated with cotrimoxazole had a higher risk of fever, vomiting, diarrhoea and positive blood smears than the chloroquine and penicillin treatment group, although the difference was not statically significant. The two cotrimoxazole treated children who were smear positive had *vivax* malaria (Table 5).

Discussion

Malaria transmission in the study area is perennial with peak incidence at the end of heavy rains from June to August. In the study year, lower average rainfall than previous years was associated with a low prevalence of positive smears of 23.1%. *P.vivax* was found to be the predominant species, accounting for 65% of cases detected. Radiographic pneumonia, on the other hand, was recorded in a larger proportion (25.8%) of children enrolled in the study than might have been expected based on data from previous years.

Health institutions in the district treat cases of pneumonia and malaria empirically based on history and physical examination. Study findings suggest that at least 13% of those treated with antibiotics suffer from untreated malaria and 15% of those treated for malaria have undetected and untreated pneumonia.

A study in Malawi (9) evaluated children under five years of age with cough or fever brought to the outpatient department of Kamuzu Central Hospital in Lilongwa. Of 1599 enrolled children, 566 (35%) had parasitaemia and 116 had radiographic evidence of pneumonia; 43 had both radiographic pneumonia and parasitaemia. Of the 471 children who met the clinical definition for pneumonia, 449 (95%) also met the malaria clinical definition. Among children with radiographic evidence of pneumonia, the clinical definition for malaria was not predictive of parasitaemia (sensitivity 93% and specificity 5%). Whether malaria parasitaemia was present or absent, the pneumonia clinical definition distinguished children with and without radiographic evidence of pneumonia (sensitivity and specificity > 60%). Children who satisfied the pneumonia clinical definition were more likely to have radiographic evidence of pneumonia (OR = 10.4, 95% CI: 5.2-20.7), parasitaemia (OR = 1.6, 95% CI: 1.2-2.2) or both at same time (OR =4.2, 95% CI: 2.1-8.4) than were children who did not meet the clinical definition.

Table 4: Response to treatment in the first week of follow up, Awassa (southern Ethiopia) 1993.

	GROUP-1	GROUP-2	Relative Risk (95% CI)
Number of children	42	49	
1.Fever	8 (19.0)	5 (10.2)	1.87 (0.66-5.27)
2.Cough	2 (4.8)	5 (10.2)	0.47 (0.10-2.28)
3.Vomiting	1 (2.4)	5 (14.3)	0.23 (0.03-1.92)
4.Diarrhoea	4 (9.5)	7 (14.3)	0.67 (0.21-2.12)

5.Temperature Febrile	6(14.3)	4 (8.2)	1.75 (0.53-5.79)
6.Fast breathing	1(2.4)	0	= =
7.Smear Positive	(2.4)	3 (6.1)	0.39 (0.04-3.60)
8.Condition			
Same	1 (2.4)	3 (6.1)	
Improved	4	1	46 0.39 (0.04-3.60)

Treatment in group-1 = Cotrimoxazole

group-2 = Chloroquine + Procaine

In the Gambia (10) a community based study of respiratory diseases in the eastern region monitored all under-five children in seven rural villages for respiratory complaints and febrile illness. Children were enrolled in the study if they had rapid respiratory rate and based on the mothers subjective impression that the child was ill. Those children with cough or fever but no fast breathing and children with chest indrawing but no fast breathing were not enrolled.

The study showed that 775 children (99.0%) satisfied the clinical case definition of malaria and 665 (84.9%) children satisfied the clinical case definition for pneumonia. Of those who met the clinical case definition for pneumonia 659 (99.0%) also satisfied the clinical case definition of malaria. Of 134 children with radiographic sign of pneumonia and negative blood films, 129 (97.0%) satisfied the clinical case definition for pneumonia. Of 107 children with radiographic signs of pneumonia and a positive blood film, 91 (85.0%) satisfied the clinical case definition for pneumonia. However, of 410 children with clinical and laboratory diagnosis of malaria but with negative radiographs, 326 (79.5%) also satisfied the clinical case definition of pneumonia. Parasitaemia was present in 520 (66.4%) of the 783 children with raised respiratory rate, and in 418 (62.9%) of the 665 who satisfied the clinical case definition of pneumonia. Only 107 (44.4%) out of 241 children with radiographic pneumonia had parasitaemia whereas 413 (76.2%) of 542 children with no radiological evidence of pneumonia had parasitaemia. Malaria parasitaemia of 5000/ mm³ and over was associated more often with abnormal chest x-ray findings in this group of children than would be expected by chance.

Like the Malawi and Gambia studies, our study showed a significant proportion of overlap in pneumonia and malaria case definitions. The overlap in clinical definitions of pneumonia and malaria of over 60% is lower than that found in the Malawi and Gambia studies. This difference could be due to the lower rate of parasitaemia and predominance of *vivax* malaria in this study when compared to the other studies. The overlap in laboratory diagnosis of both parasitaemia and radiographic pneumonia was similar to that found in the Mozambique study.

In contrast to the study in Malawi, our investigation revealed no reduced incidence of parasitaemia in the presence of radiographic pneumonia. In addition, radiographic pneumonia was more often detected in this study, though not in Malawi, among those who met the clinical case definition for pneumonia when the smear was negative than positive. But the findings of this study were similar to those in the Gambia, where radiographic pneumonia was more often detected among those who met the clinical case definition of pneumonia when they were smear negative than when they were smear positive.

However, the overall likelihood of radiographic pneumonia was higher for smear positive than smear negatives. This relationship was unaffected by differences in age, sex, haemoglobin, or parasite type and density. This result contrasts with findings of previous studies.

Table 5: Response to treatment in the second week of follow up, Awassa (southern Ethiopia) 1993.

	GROUP-1	GROUP-2	Relative Risk (95% CI)
Number of children	41	46	
1.Fever	3 (7.3)	2 (4.4)	1.68 (0.30-9.58)
2.Cough	0	1 (2.2)	= =
3.Vomiting	2 (4.9)	0	= =
4.Diarrhoea	4 (9.8)	3 (6.5)	1.50 (0.36-6.29)
5.Temperature Febrile	2 (4.9)	2 (4.4)	1.12 (0.17-7.61)
6.Fast breathing	0	0	= =
7.Smear Positive	2 (4.9)	0	= =

8.Condition			
Same	2 (4.9)	0	
Improved	39	46	= =

* Treatment in group-1 = Cotrimoxazole
group-2 = Chloroquine + Procaine

A facility-based study in Nigeria (8) evaluated the risk of malaria with bacteraemia in preschool children. Six hundred and forty two febrile preschool children without localizing signs were found to have a prevalence of bacteraemia which was comparable in those with malaria (43/446, 9.6%) to those without malaria (24/196, 12.2%, $p < 0.5$). In Mozambique a study was conducted among 220 children between two, four months and four years of age admitted with the diagnosis of pneumonia (11). Of 220 children with pneumonia, 19 (8.6%) also had *P.falciparum* parasitaemia. Among those with parasitaemia 12 (63%) were classified as having severe pneumonia. The 19 children with pneumonia and parasitaemia had a longer mean duration of cough and fever before admission (5.5 days compared to two days).

The study in Nigeria showed a higher prevalence of both malaria and bacteraemia among anaemic infants while the study in Mozambique documented lower haemoglobin levels among children with pneumonia and malaria. In the study in the Gambia, chest x-ray changes were associated with higher parasite densities while in Nigeria, no correlation was observed between parasite density and overlap of pneumonia and malaria (8-11).

The specificity of the clinical definitions of malaria and pneumonia were not adequate to differentiate between the two diseases. Inclusion of other additional criteria to improve the specificity of the clinical definition might reduce over treatment, but would still not permit pneumonia to be distinguished clinically from malaria. The presence of a palpable spleen in malaria and crepitation in pneumonia increased the predictive value of the case definitions, but these findings were present in only a small proportion of the cases. This study was similar to previous studies in finding a low predictive value of fever in pneumonia, while maternal reports of fast or difficult breathing were better predictors of the presence of pneumonia.

Cotrimoxazole was found to be as effective in the treatment of malaria as chloroquine. All patients in both groups had a statistically significant response. Parasite clearance rates were better in the cotrimoxazole than the chloroquine group, however the cotrimoxazole group had the highest rate of recurrences. This finding is similar to that of the Tanzania and Mozambique studies, which showed a higher rate of recurrence with cotrimoxazole despite an effectiveness in parasite clearance equal to that of chloroquine.

The chloroquine group was less likely to have persistent fever, though more likely to have gastrointestinal complaints. Further studies are needed to assess chloroquine resistance in the area.

In conclusion, the study shows a significant proportion of overlap in the clinical and laboratory diagnosis of pneumonia and malaria shown and that a three-day cotrimoxazole therapy is equally effective in treating both diseases as a combination of chloroquine and procaine is.

Since, the overlap in the clinical presentations is unaffected by the type of *plasmodium* and the risk for malaria and pneumonia are similar throughout the country, the results of this study can be generalized within Ethiopia to assist clinical management of children under 5 years old age.

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Original article

Tuberculosis and HIV infection in southern Ethiopia

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Abstract: A health institution-based cross-sectional study was carried out in Shashemene town, southern Ethiopia, between September, 1993 and January, 1994 to determine the sero-prevalence and the clinical impact of HIV among newly diagnosed tuberculosis patients. The HIV-antibody was determined using the two ELISA procedures (Wellcozyme, Wellcome Diagnostics, Dartford Kent England, and Du-pont assay, Singapore). A total of 450 tuberculosis patients aged 15 years and above were enrolled in the study. The overall HIV-seroprevalence rate was 44.4%. The highest rate was observed in the age group 20-39 years. A slightly higher HIV-infection rate was found in males (46%) than in females (41%). Those divorced and widowed patients had higher proportion of HIV sero-positivity. The HIV positivity rate was higher for extra-pulmonary than pulmonary form of tuberculosis (OR = 3.80; 95% CI: 1.49, 9.7). Higher proportions of sputum positive patients were HIV-positive compared to the sputum negative pulmonary tuberculosis patients (OR=1.09; 95% CI: 0.64,1.85) though they manifested typical radiographic features (OR = 7.87; 95% CI: 4.39, 14.21). Significant differences were noted among HIV positives than HIV negatives in manifesting herpes zoster, lymphadenopathy, oral candidiasis, peripheral paraesthesia and chronic diarrhoea. An alternative diagnostic approach is required to avoid missing HIV-related tuberculosis which is communicable to the general population. Other findings are discussed and recommendations made. [*Ethiop. J. Health Dev.* 1997;11(1):51-59]

Introduction

Tuberculosis remains a health problem of enormous dimensions, particularly in the developing world, affecting millions of people each year. The pandemic of the acquired immunodeficiency syndrome (AIDS) and the evidence of an association between TB and HIV, which causes AIDS is now a further cause for world-wide concern (1-4).

Considering that the great majority of HIV infections occur among the 15-49 year-old,(5,6) and assuming that the risks of infection with HIV and tuberculosis are independent, it has been estimated that world wide more than 4 million persons, again mostly in the developing countries, have been infected with both HIV and *Mycobacterium tuberculosis*.

The most important risk factor yet identified for the development of tuberculosis following infection is human immunodeficiency virus infection. Very high incidence rates of TB in dually infected individuals have been reported in both industrialized and developing countries (7).

In the developing countries, the overlap between HIV infection and tuberculosis is shown by the high HIV sero prevalence among patients presenting with active tuberculosis. Data from a number of African countries and Haiti show that sero-prevalence ranges between 17% and 66% (2, 8-12). Developing countries are also faced with the problem of the increased cost of tuberculosis treatment brought about by the increase in numbers of cases (13).

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The HIV pandemic will worsen the TB situation in developing countries in three ways over and above the existing situation by (a) reactivation of a latent TB infection among dually infected persons (b) new infection with tubercle bacilli and rapid progression to active disease in HIV infected persons and (c) increasing the number of cases in the general population whose infection and disease is the result of transmission from HIV positive individuals developing TB by either reactivation or recent infection (14-16).

The influence of HIV-infection on the clinical picture of tuberculosis has been examined by studies in Africa (2) and elsewhere (17,18). In Ethiopia one study, conducted among 106 soldiers in 1988, has reported a 6.6% seropositivity among TB patients (19). Although a 1990 national tuberculin survey has shown an annual tuberculosis infection rate of 1.4%, and the disease is ranked first as cause of death for adults admitted to health care units (20), its relationship to HIV infections has not been well studied. The present study was thus undertaken to assess the magnitude of HIV infection in tuberculosis patients and describe the clinical manifestation of tuberculosis in HIV infected individuals.

Methods

This health institution-based cross-sectional study was conducted in Shashemene, a town located in southern Ethiopia, between September, 1993 and January, 1994. The town is found 250 kms along the main road from Addis-Ababa to Moyale-Kenya. It has a wide communication network connecting more than four regions of the country.

The sources of the study subjects include both inpatient and outpatient departments of Shashemene General Hospital, one Health Centre and another two health institutions (Gambo Catholic Hospital and Awassa Health Centre) which frequently refer patients to the Shashemene Hospital for the anti-TB treatment. All new tuberculosis patients who came seeking the services of the health institutions were included in this study. New patients with a suspected and/or confirmed diagnosis of tuberculosis between September 5, 1993 and January 10, 1994 at the local hospital and health centre were entered in the study.

The purpose of the study was explained to the heads of health institutions in the study areas and to the clinicians who work there. Physicians in the health centres and hospitals were asked to refer all suspected TB cases to the research team for further investigation of the patient. Accordingly a total of 450 newly diagnosed TB patients were enrolled in the study.

To maximize efficient utilization of the ELISA reagent, patients' sera were stored at 20°C in Shashemene General Hospital and later analysed according to the Kit manufacturer's instructions.

The following operational definitions of tuberculosis were used: *confirmed TB diagnosis*: Pulmonary TB was diagnosed when there are two Acid Fast Bacilli(AFB) positive results of sputum specimen; or a patient with one sputum specimen positive for AFB and radiologic abnormalities consistent with active pulmonary tuberculosis. *Suspect case*: patient in whom presumptive diagnosis is made on the basis of radiological or other clinical evidence and with negative AFB smear. *New case*: patient who was diagnosed with tuberculosis for the first time. *Disseminated TB*: there are more than two organs, other than pleural and lymph node, affected by the lesion. *HIV-Positivity* was defined to occur when patients tested positive to the combination of Wellcozyme and Dupont assays.

A standardized questionnaire was used to collect demographic and clinical information. Physicians from each health institution filled out the questionnaires. In this questionnaire, information relating to their history and clinical examination was completed for each patient, radiological and laboratory results were recorded and blood samples taken by laboratory technicians. All patients with pulmonary tuberculosis had a chest X-ray. Radiological features, including the zones involved and the presence or absence of cavities, were recorded by the physician at the time of recruitment.

Seroprevalence was determined for all new TB patients who were 15 years of age and above, irrespective of the lesion site. All patients with suspected/confirmed TB were enrolled for the

analysis of clinical presentations. Patients seen at the health institutions were treated according to the national guidelines by the respective health units. Serum samples were analysed for the HIV antibody by a competitive recombinant enzyme linked immunosorbent assay (Wellcozyme, Wellcome Diagnostics, Dartford, Kent UK) and antiglobulin recombinant ELISA assay (Dupont, Singapore). There are as yet, no reports of HIV-2 in Ethiopia. Throughout this text HIV-1 seropositivity is referred to as HIV-seropositivity.

Procedures were performed according to the kit manufacturer's instructions. Only those sero-positive on both assays were considered HIV-positive. Studies in Addis Ababa (21), Kenya (22), and Zambia (2,23) showed this definition to be concordant with Western-Blot for HIV-1. This examination strategy is also supported by a recent WHO report (24). Sputum was examined for acid fast bacilli on direct smear using Ziehl-Neelsen stain and fluorescence microscopy with a 25 x objective and 6.5 x eye pieces. Serological testings were carried out anonymously, with all clinical and laboratory data being identified only by a code number.

A chest X-ray was performed for all pulmonary tuberculosis patients and read by three general practitioners (with similar year of experience) who have undertaken special training on the radiology of the chest to run the tuberculosis follow-up clinics.

Data entry and analysis were done using EPI INFO and SAS computer statistical packages. Initial analysis of the association between HIV seropositivity and various variables were performed using standard 2 x 2 tables. Further analyses were carried out using logistic regression. Where appropriate, Maentel Haenszel Chi-square methods were applied to test for trends. The odds ratios (OR) presented are the antilogs of the logistic regression co-efficient. Ninety-five percent confidence intervals were calculated and statistical significance was designated at $P < 0.05$.

Patients were informed of the study procedures and the tests to be made during the investigation. Informed verbal consent was obtained from each participant. TB treatment was free for all study participants and anti-TB drug stock adequate for full course was secured for both study centres. Counselling service to be offered to HIV positive patients was organized in collaboration with the Zonal Health Department prior to the initiation of the study. The study was approved by the ethical committee of both the Department of Community Health and Faculty of Medicine, Addis Ababa University.

Results

Between September, 1993 and January, 1994, a total of 450 tuberculosis patients were enrolled in the study. Of the 450 patients there were 269 (60%) males and 181 (40%) females. The mean age of the patients was 30 years. Most patients 254 (56%) were from Shashemene, 97 (22%) were students. Farmers and housewives accounted for 17% each. The remaining 203 (45%) belonged to different other occupations. (Table 1).

Overall, 199 (44%) of the patients were positive for HIV. There were nine indeterminate results who were negative by Wellcozyme and positive by the Du-pont assay. These patients were considered as HIV negatives.

As indicated in Table 1, there was no statistical difference in HIV sero-prevalence with regard to age and sex. However, in the age group of 20-39 and in male patients HIV positivity was found to be higher. The peak sero-prevalence occurred in the age group 20-29 years. Sex was not associated with HIV status in these patients when adjusted for age. The adjusted odds ratio (OR) and 95% confidence interval (95% CI) were 0.84(0.57, 1.21).

There was a statistically significant association between marital status and HIV positivity. (OR=0.60, 95% CI :0.40,0.88 $p<0.05$).The association persisted even after adjusting for age and sex. Patients who were widowed/separated or divorced had higher prevalence of HIV positivity. Occupational categories which were significantly associated with HIV positivity were government employees, prostitutes, merchants, ex-soldiers, and daily labourers.

Table 1: The socio-demographic distribution of HIV positives among tuberculosis patients, southern Ethiopia, 1994.

HIV Status	
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Age	Positive	Negative	Adjusted OR (95% CI)
15-19	27(40.9)	39(59.1)	1 ²
20-29	76(47.8)	83(52.2)	1.48 (0.86, 2.56)
30-39	62(46.6)	71(53.4)	1.56 (0.92, 2.64)
≥40	34(36.9)	8(63.1)	1.18 (0.61, 2.25)
Sex			
Male	24(46.1)	45(53.9)	0.84 (0.57, 1.2)
Female	75(41.4)	06(58.6)	1.00 ²
Marital Status			
Single	00(50)	03(41)	1 ²
Married	79(40)	35(54)	0.60 (0.40, 0.88)
Other	20(10)	13(5)	1.56 (0.73, 3.3)
Occupation			
Farmer	25(33)	50(67)	1 ²
Student	32(33)	65(67)	1.98 (0.51, 1.86)
Driver	9(35)	17(65)	1.05 (0.40, 2.7)
Hous-wife	27(36)	48(46)	1.12 (0.57, 2.2)
Daily labourer	13(52)	12(48)	2.15 (0.85, 5.4)
Ex-Soldier	26(55)	21(45)	2.46 (1.16, 5.2)
Merchant	22(59)	15(41)	2.91 (1.28, 6.59)
Prostitute	9(60)	6(40)	2.97 (1.94, 3.4)
Gov't employee	23(64)	13(36)	3.50 (1.50, 8.1)
Other	13(76)	4(24)	6.5 (1.92, 22.1)
	27(40.9)	39(59.1)	1 ²

¹Adjusted odds ratio and 95% confidence interval. The logistic regression model included age (4 levels), sex, marital status (3 levels) and occupation (10 levels).

²Referenc category

Two hundred sixty nine patients were diagnosed as having pulmonary disease on the basis of clinical findings, radiographic and/or sputum smear results. In 128 (48%) of them tubercle bacilli were identified by the Ziehl Neelson stain method. In this study, patients with extrapulmonary tuberculosis were more likely to be HIV sero-positive than were patients with pulmonary lesions. Table 2 shows the relationship between HIV status and site of tuberculosis. Extrapulmonary disease (pleural, lymph node, disseminated), both alone and in combination with parenchymal lung disease, was strongly associated with HIV infection (OR = 3.92; 95% CI: 1.54, 9.9) $P < 0.05$. The association persisted when adjusted for age and sex (OR = 3.80 95% CI: 1.49, 9.7) $P < 0.05$. The lungs were, however, the commonest site of the disease in both HIV positive and negative patients.

The overall proportion of hospitalized tuberculosis patients was 29%. Out of these 41% were positive for HIV infection, while 46% of the ambulatory TB patients were sero-positive. The sero-prevalence of HIV infection in hospitalized TB patients was not significantly different from those who attended the out-patient department. No significant association between HIV status and sputum examination result for AFB was observed in this study.

Table 2: Association between sites of tuberculosis and HIV sero-positivity, southern Ethiopia, 1994.

	HIV Status			OR (95% CI)
	Positive N=199	Negative N=251		
Pulmonary	95	174	1 ¹	1 ²
Pleural	12	6	3.66(1.33, 10.0)	3.45(1.24
Disseminated	28	27	1.89(1.05, 3.40)	2.02(1.11
Lymph node	49	37	2.42(1.47, 3.97)	2.48(1.40
Pulm + EP	15	7	3.92(1.54, 9.9)	3.80(1.49

¹Adjusted for age and sex by logistic regression.

²Reference category

Chest X-rays were available for all pulmonary TB patients (Table 3). The chest X-rays showed pulmonary cavitation in 79 (55%) of the HIV negative, but in only 65 (45%) of those HIV-positives. (OR = 2.61; 95% CI: 1.49, 4.56, $P < 0.05$). In seventy six (52%) of the 174 HIV negative pulmonary TB patients upper zone lesions predominated compared with 70 (48%) among the 95 HIV positive cases. (OR = 3.61; 95% CI: 2.02, 6.48, $P < 0.05$). When the association between HIV status and AFB sputum smear were positivity stratified for the presence of cavitation and superior segment involvement, trends were still present, but were less marked and were not statistically significant.

As shown in Table 4, patients who had upper lobe infiltration appear to be nearly equally divided between sputum smear positives and negatives; and most sputum positives appeared to have

pulmonary cavitation (OR = 7.87; 95% CI: 4.39, 14.21).

Table 3: Radiographic findings in cases with pulmonary tuberculosis in relation to HIV infection, southern Ethiopia, 1994.

X-ray findings	HIV Positive n=95	No (%)	HIV Negative n=174	No (%)	Adjusted ² OR (95% CI)
Upper zones only					
Yes	70(48)		76(52)		1.00 ²
No	25(20)		98(80)		3.61(2.02, 6.48)
Middle/Lower Zone					
Included	35(30)		80(70)		1.00 ²
Not Included	60(39)		94(61)		0.69(.40, 1.18)
Cavitation					
Present	65(45)		79(55)		1.00 ²
Absent	30(24)		95(76)		.61(1.49, 4.56)

¹Adjusted for age and sex by logistic regression.

²Reference category

Table 5, shows the relationship between symptoms and signs at presentation and HIV status. A significant number of sero-negative TB patients had complained of cough and fever. Prolonged history of cough and fever was also reported by a large proportion of TB patients. There was no significant difference in the history of marked weight loss and organomegaly among the HIV positive and negative patients. A large proportion of patients who presented with lymphadenopathy were positive for HIV.

Clinical signs and symptoms which showed an association with HIV infection were generalized lymphadenopathy, oral candidiasis, herpes zoster, chronic diarrhoea and paraesthesia. Sputum positive tuberculous patients showed a slight difference from sputum negatives in the development of fever. Most sputum negative patients had a more prolonged fever than the others. Large numbers of sputum negative patients tended to manifest with chronic cough than smear positive patients ($\chi^2 = 29.15$; df = 2, $P < 0.001$).

Discussion

The sero-prevalence rate of 44.4% in this study is consistent with other studies conducted in Sub-Saharan Africa, the Caribbean and in some urban areas of the United States that have shown 20-60% of tuberculosis patients to be HIV sero-positive (3,4). However this rate is much higher than that previously reported from testing of 106 Ethiopian soldiers in 1990

(19). This marked difference could be attributed to study size differences and to the rapid dissemination of HIV infection over time, with essential consideration of the time of introduction of the virus in the country and the level of immune compromise. Although evidence on the proportion of HIV/AIDS patients who developed tuberculosis is required this study partly supports the new HIV/AIDS case definition for Ethiopia which considers TB as a major criterion.

Table 4: Association between sputum smear positivity and radiographic appearances among pulmonary TB patients, southern Ethiopia, 1994.

	Sputum for AFB		Adjusted ¹ OR (95% CI)
	Positive	Negative	
Cavity			
Present	00(69)	44(31)	1 ²
Absent	28(22)	97(78)	7.87(4.39, 14.21)
Upper zone involvement			
Yes	52(52)	48(48)	1 ²
No	76(44)	93(56)	.33(0.78, 2.24)

¹Adjusted for age and sex by logistic regression.

²Reference category

The findings in this study are also in conformity with the other observations that have been noted in the USA, Europe and Africa (1,2,5-12).

The higher sero-positivity observed in this study among the age group 20-39 years is comparable with other African studies (2,6,8,13). The younger ages at which sero-reactive persons developed tuberculosis probably reflects the age prevalence of HIV sero-positive persons in the community, as it is similar to that reported from the surveillance data in this country (21).

Distribution of HIV positivity across the various occupatons in this study indicates the wider distribution of the infection in the community. Accordingly, the practice of concentrating on specific groups of the population while addressing health education deserves re-consideration.

The association of HIV infection with marital status is compatible with the known transmission dynamics of HIV. Being single has a higher risk of acquiring the infection.

Table 5: HIV sero-positivity and selected clinical findings among tuberculosis patients, southern Ethiopia, 1994.

Clinical Findings	HIV +ve No (%) (n=199)	HIV -ve No (%) (n=251)	Adjusted ¹ OR (95% CI)
Candidiasis			
Present	12(6)	5(2)	3.16(1.02, 10.4) 0.02
Absent	187(94)	246(98)	1 ²
Lymphadenophaty			
Absent	58(29)	51(20)	1.61(1.02, 2.55) 0.03
Present	141(71)	200(80)	1.00 ²
Herpes Zoster			
Absent	14(7)	6(2)	3.46(1.21, 10.3) 0.008
Present	185(93)	245(98)	1.00 ²
Hepatomegaly			
Present	35(18)	52(21)	0.82(0.49, 1.35) 0.40
Absent	164(82)	199(79)	1.00 ²
Splenomegaly			
Present	164(15)	199(79)	1.22(1.74, 2.03) 0.40
Absent	35(85)	52(21)	1.00 ²
Paraesthesia			
Present	30(15)	17(7)	2.44(1.25, 4.80) 0.004
Absent	164(85)	234(93)	1.00 ²
Marked weight loss			
Present	124(62)	155(62)	1.02(0.69, 1.53) 0.09
Absent	75(38)	96(38)	1.00 ²
Fever			
None	8(4)	10(4)	1.00 ²
< one month	67(34)	92(36)	0.91(0.34, 2.4) 0.5
≥One month	124(62)	149(59)	1.04(0.39, 27)
Cough			
None	40(20)	40(16)	1.00 ²
<one month	35(18)	42(17)	0.83(0.44, 1.56) 0.45
≥one month	124(62)	196(67)	0.73(0.44, 2.04)
Diarrhea			
None	53(77)	221(88)	1.00 ²
<one month	32(16)	18(7)	2.56(1.39, 4.74) 0.008
≥one month	14(7)	12(5)	1.68(0.75, 3.74)

¹Adjusted for age and sex by logistic regression.

²Reference category

The sero-prevalence of HIV-infection in hospitalized TB patients was not significantly different from those attending the out-patient department. However, inferences regarding the prevalence of HIV infection in hospitalized TB patients depends on many factors that may have changed over

time, such as HIV specific mortality rates and criteria for hospital admission and discharge. Examination of this variation between the two sources of TB patients is required through further study.

In this study 29% of the patients with tuberculosis were admitted to the hospital for anti-TB treatment. This indicates the effect of HIV on work load of hospitals. The impact of HIV on resource utilization like hospital beds, diagnostic investigations and therapies has been studied elsewhere (16). This also reinforces the importance of educating all personnel involved in health care to take precautions against occupational exposure to HIV with all patients and to avoid possible nosocomial infections. In the absence of rapid and effective interventions, increasing numbers of HIV/TB cases are likely to occur and affect hospital admissions in Ethiopia.

In this study the lung was the commonest site of disease in both HIV positive and negative patients. Only 23% of HIV positive patients had positive sputum smears. Even though the absolute number of 46 was much less than the number of sputum-positive HIV negative cases, the role of HIV for the spread of the disease in the community should not be underestimated. Since pulmonary disease is probably more important in the transmission, the contribution of HIV positive TB patients to the spread of the disease is likely to be greater despite this association of HIV with extrapulmonary disease.

The proportion of HIV positive TB patients who had extrapulmonary infection alone (35%) was similar with that reported in a cohort study in Zambia (23), but was higher than other studies done in Zaire, Haiti and Ivory coast (2,11,12). This difference could have resulted from bias towards pulmonary disease during recruitment in those studies which were conducted at TB referral centres. Lymphadenopathy remains the most frequent form of extrapulmonary TB as reported from Central Africa Republic, Kenya, Zambia and Brazil (6). A study from Zaire also showed that patients with suspected TB and with extrapulmonary TB had higher HIV seropositivity rates than patients with sputum confirmed TB (2). This fact should be seen in the light of an essential consideration; i.e., a general shift of trend from pulmonary to extrapulmonary tuberculosis in the present HIV/AIDS era, thus leading to diagnostic difficulties by challenging/impeding the available diagnostic possibilities.

Similar to other evidences obtained (2,11,22), there was no appreciable difference in HIV seropositivity among sputum smear positive and negative pulmonary TB patients. Similar findings were reported elsewhere (2,11,22), although others have reported a tendency for HIV positive patients to have a negative sputum smear (9). This variation may account for the level of immuno-deficiency posed by HIV infection and prior exposure to TB. The negative sputum smear among HIV positive patients probably indicates that HIV positive TB patients may frequently have dry cough. The TB detection rate based on sputum smear examination may thus be comparable to that described for HIV sero-negative patients, at least during the early stages of HIV disease (6).

The classic form of TB, with cavitating, upper zone, pulmonary lesions, is known to be determined by the interaction between the bacillus and the host's immune system so that, in the presence of immuno-deficiency, a different outcome may be expected (8).

In this study pulmonary cavitation was observed more often in HIV sero-negative TB patients when compared with HIV sero-positive TB patients. This finding is incompatible with other reports (2,6), and could be explained by the low grade immuno-depression in the study subjects, leading to the typical cavitory lesion. Similar findings have been reported (26,27) attributing the lesion to the interaction between the bacillus and the host's immune system.

In contrast to other evidence elsewhere (6,26,27) this study revealed that most HIV negative patients with pulmonary tuberculosis tend to have the classic upper lobe involvement. However, a large proportion of HIV positive patients had atypical presentation having supplementary middle/lower zone involvement and non cavitory appearance. These findings further strengthen the distorted radiographic appearance of TB patients which was established elsewhere (6).

A discrepancy between pulmonary cavitation, sputum positivity and upper lobe involvement was observed in this study. All sputum positive cases did not show cavitation and /or upper segment

involvement. It has been observed that even in the absence of cavitation the profound impairment of the immunity seen in HIV infection may permit tubercle bacilli to multiply in such large numbers as to become visible on smear examination (11). Therefore the above finding could probably be a reflection of this phenomenon.

Both tuberculosis and HIV infection lead to chronic ill-health and wasting, and both are associated with persistent cough and fever. These characteristics were also seen in this study, illustrating the difficulties in clinical diagnosis. Patients with tuberculosis, with or without HIV infection, might be diagnosed as having HIV/AIDS alone and might therefore fail to receive the anti-TB treatment.

This study examined the impact of HIV on the process of diagnosis of TB. Symptoms of TB were comparable in both groups. HIV positive and negative patients, had similar complaints of cough, although fever was more frequent and of longer duration in HIV positives.

The presence of generalized lymphadenopathy, peripheral paraesthesia, herpes zoster, oral candidiasis and diarrhoea may assist in identifying patients with dual infection with tuberculosis and HIV but do not contribute to the diagnosis of TB itself. The lack of association between HIV and prolonged diarrhoea in this study could be attributed to possible recall bias. The tendency of sputum smear negative patients to manifest with prolonged cough and fever further threatens the sputum detection rate, thus compounding the difficulty of clinical diagnosis of pulmonary tuberculosis.

In the absence of a quick and cheap diagnostic test, physicians may be obliged to treat patients with prolonged fever, cough, weight loss, negative sputum smear for acid fast bacilli and failure to respond to standard antibiotics, as tuberculosis cases without confirmation of the diagnosis.

It is worth mentioning that this cross-sectional study lacks information about the role of HIV infection on tuberculosis relapse. A longitudinal design is required to address this issue.

It is to be noted that there are certain limitations to the study. The nature of the study itself has limited the collection of complete information from the patients regarding the clinical signs and symptoms. Nevertheless, since all new tuberculosis patients seen at the health institutions during the study period were included in the study, and the physicians involved in clinical examination of patients were the same, it is assumed that in general, the information obtained was comparable for all patients. Confirmation of TB diagnosis was not possible because of the inherent limitation of the diagnostic tests. Lymph node biopsy could not be done for logistic reasons. But it is unlikely that the limitations pointed out would compromise the main findings of the study.

The urban-rural mix of the study subjects and uniformity of the selection criteria used for enrolling new TB patients with adequate sample size permits the internal validity and wider generalizability of the study findings.

In conclusion, the results show a high prevalence of HIV infection among tuberculosis patients. The results also show the difficulties of clinical diagnosis of co-infected patients who manifested unusual clinical, laboratory and radiographic features. A diagnostic approach with a higher degree of diagnostic ability is required to avoid missing HIV-related tuberculosis which is communicable to the general population.

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Original article

Effect of natural fermentation on nutritional and antinutritional factors of tef (*Eragrostis tef*)

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Abstract: Tef flour mixed with water in a 1:1.6 (w/v) ratio was allowed to ferment at 22^o C for 96 hrs by the action of endogenous microflora in the batter. After 96 hrs total protein content in tef dough decreased by 12% whereas the NPN, free amino acids, free amino acid nitrogen, soluble protein and fat acidity increased 7.4-, 7.0-, 6.6-, 7.7- and 10.7- fold, respectively. Fermentation

also resulted in significant drop in pH and sharp rise in titratable acidity of the 96 hrs fermented dough. Iron, phosphorus and calcium decreased by 43%, 35% and 41%, respectively, in the dough fermented for 96 hrs. Phytic acid, tannins and trypsin inhibitor contents were reduced by 72%, 55% and 69%, respectively. In *ersho*, the liquid portion drained off from the fermented dough, total protein, NPN, free amino acids, iron, calcium and phosphorus increased significantly whereas total and reducing sugars decreased during the two days of fermentation. The pH of *ersho* dropped slightly but the titratable acidity increased by 35%. These results could provide useful indices for the improved evaluation of tef fermentation. [Ethiop. J. Health Dev. 1997;11(1):61-66]

Introduction

Tef (*Eragrostis tef*) is the second most extensively cultivated crop indigenous to Ethiopia (1). Tef is commonly used in Ethiopia in the production of beverages and a number of foods, such as *injera* (leavened pancake), *kita* (unleavened pancake), porridge and gruel (2). *Injera* is a pancake-like soft, thin leavened bread obtained by natural fermentation of cereal grains including tef. The fermentation process lasts for a total of two to four days. Sequences of events and successions of microorganisms during the fermentation of tef have been previously reported (3,4). Tef *injera* contributes a major part of the diet for the Ethiopian population with the exception of those living in areas where Enset (*Ensete ventricosum*), maize and sorghum are the main diets. *Injera* is eaten with stew prepared from legumes or meat.

Like other cereals, tef may contain considerable amount of antinutritional factors like, phytic acid, tannins and trypsin inhibitors. Phytic acid makes iron, calcium, zinc and magnesium unavailable for use by the body and also binds with proteins (5). Tannins might also reduce protein digestibility by inhibiting the digestive enzymes (6). High levels of trypsin inhibitor activity stimulate pancreatic juice secretion and cause pancreatic hypertrophy and growth (7).

Fermentation has been reported to significantly decrease phytic acid, tannins and trypsin inhibitors (8) and improve protein digestibility (*in vitro*) and the nutritive value of cereals and legumes (9,10). The effect of natural fermentation on antinutrients in tef is, however, lacking. On the other hand, fermentation processes negatively affect the chemical composition and the overall nutritive value of a diet (9,11). A substantial loss of protein (40%) was reported during the fermentation of maize and millet to *ogi* (12). Fermentation of enset also resulted in loss of protein, carbohydrate and calcium (13,14). Natural fermentation has also been reported to decrease the total

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nitrogen content of tef dough (3). *Erscho*, the clear yellow liquid that accumulates on the surface of the fermenting tef-flour batter is discarded during *injera* preparation. The microbial flora and chemical properties of *erscho* has been reported (15). There is, however, paucity of data indicating the loss of nutrients during tef dough fermentation. The present communication deals with studies on the levels of antinutritional components and loss of nutrients during tef dough fermentation.

Methods

White variety tef (*Eragrostis tef*) was purchased from an open market in Addis Ababa, Ethiopia. The seeds were cleaned by sieving to remove foreign matter. The cleaned tef was ground through a 0.5 mm screen in Cyclotec sample mill (Tecator, Sweden). Dough was prepared by mixing the flour with water in a 1:1.6 (w/v) ratio and homogenized. The homogenous slurry was allowed to ferment at 22^o C for 96 hrs in a glass jar covered with aluminium foil. Samples were removed at the beginning of fermentation, and thereafter at 24 hr intervals, for pH determination and chemical analysis for up to 96 hrs. *Erscho* was collected and its volume measured. To measure the pH of the dough, 10g of the fermenting mass was suspended in equal volume of deionized water and homogenized. pH of the suspension was measured using an Orion (Orion, USA) pH meter. The

titratable acidity expressed as percent lactic acid was determined by titrating the decanted homogenate used for pH measurement against 0.1 NaOH to pH 8.3 end point. pH and titratable acidity of *ersho* was determined without further treatment.

For chemical analyses, the fermented dough was dried at 60°C in an oven. The dried, fermented dough was reground in the Cyclotec sample mill and stored at 4°C until analyzed.

Total protein of the dough and *ersho* was calculated using the factor 6.25xN after determination of total nitrogen by micro-Kjeldahl method described in the Association of Official Analytical Chemists (AOAC) (16). Total sugar in *ersho* was estimated colorimetrically according to Dubois *et al* (17) and reducing sugars by the method of Miller (18). The difference between total and reducing sugars was taken as non-reducing sugars.

The method Vanillin-HCl described by Maxon and Rooney (19) was employed for the estimation of tannin in tef dough using catechin as a standard and expressed as catechin equivalents (mg/100 g, dry weight).

Trypsin inhibitor activity in tef dough was determined by the method of Kakade *et al* (11) using N-benzoyl-DL-arginine-p-nitroaniline (BAPNA) the trypsin substrate and expressed as TIU/g (dry, weight).

Total iron in tef dough and *ersho* was determined using bathophenanthrolein as described in AOAC (16), phosphorus by the Fiske-Subbarow method (20) and calcium according to the AOAC (16).

Nonprotein nitrogen (NPN) in tef dough and *ersho* was determined by the procedure of Concon and Soltess (21). The ninhydrin method of Rosen (22) was used to estimate free amino acids in tef dough and *ersho* using leucine as a standard. The degree of protein hydrolysis in the fermented tef dough and the discarded liquid portion was determined according to Tangnual *et al* (23). Fat acidity was determined in 2.0 g samples extracted with 20 ml toluene-ethanol (1:1 v/v), and titrated with 0.0178N KOH. It is expressed as mg KOH/100 g sample (24).

Data analysis: Data were subjected to analysis of variance (ANOVA). Differences were considered significant at $p < 0.05$.

Results

Table 1 indicates changes in protein components, fat acidity and protein solubility of fermenting tef dough. As fermentation time increased total protein of the dough decreased. Total protein content of the dough decreased by about 12% after 96 hrs of fermentation.

Nonprotein nitrogen (NPN) increased significantly ($p < 0.05$) throughout the fermentation process. This fraction presumably consisted, predominantly, of the products of storage protein degradation-amino acids and peptide. The increase accounted for the loss in true protein which was not estimated in this study. The proportion of NPN relative to protein gradually increased. After 96 hrs of fermentation, more than 75% of the nitrogen was in the form of protein. The total amount of amino acid nitrogen, as a measure of protein hydrolysis, increased more than 9-fold during the 96 hrs fermentation as proteins were degraded. Free amino acids and protein solubility similarly increased nearly 7- and 8 - fold, respectively, after 96 hrs of fermentation period.

Fat acidity increased significantly ($p < 0.05$) during the first 48 hrs of fermentation which then remained unchanged throughout the fermentation period.

Table 1: **Changes in protein components, fat acidity and protein solubility of fermenting tef dough**

Period hr	Total protein %	Nonprotein Nitrogen %	Amino acid Nitrogen mg/g	Free amino Acids mg/g	Soluble Protein %	Fat acidity mgKOH/100g
0	23.10±0.10 ^a	0.11±0.01 ^a	5.73±0.13 ^a	75.3±1.2 ^a	0.41±0.03 ^a	0.11±0.01 ^a
24	22.92±0.05 ^a	0.22±0.03 ^b	26.24±0.11 ^b	359.3±3.6 ^b	4.33±0.12 ^b	0.91±0.07 ^b
48	22.42±0.13 ^b	0.53±0.02 ^c	27.93±0.08 ^b	393.4±4.7 ^c	5.41±0.09 ^c	1.15±0.04 ^c

72	21.59±0.07 ^c	0.68±0.03 ^d	30.17±0.11 ^c	424.3±7.3 ^d	5.93±0.11 ^d	1.16±0.07 ^c
96	20.33±0.11 ^d	0.81±0.04 ^e	37.60±0.12 ^d	527.2±8.7 ^e	6.97±0.13 ^e	1.18±0.06 ^c

Values are means ± S.D. of five determinations. Values in a column followed by different superscripts are significantly different (p<0.05).

The freshly prepared tef dough started to ferment soon after the addition of water and the pH of fermenting tef dough decreased significantly ($p<0.05$) from about 6.1 to 3.8 during the first 72 hrs. Thereafter, the pH of the fermented tef dough remained at similar low levels. Concomitantly with the drop in pH was a sharp rise in titratable acidity of the medium. The highest titratable acidity attained in tef dough fermented at room temperature was 1.26%.

A significant difference was observed in the nonfermented and fermented tef dough samples in their phytic acid contents (Table 2). The amount of phytic acid decreased by 72% by the 96 hr of fermentation. Similarly, tannin and trypsin inhibitor activity contents decreased significantly ($p<0.05$) by 55% and 68.9%, respectively, after 96 hrs of fermentation.

The changes in minerals content is shown in Table 2. All the minerals studied, (calcium, iron and phosphorus), significantly ($p<0.05$) decreased throughout the fermentation period. The highest decrease was observed in iron (43%) after 96 hrs of fermentation. Table 3 indicates changes in protein components and minerals in *ersho*. The total protein, NPN, amino acid nitrogen and free amino acids content increased significantly ($p<0.05$) with the fermentation time. The content of calcium, phosphorus and iron in *ersho* also increased with the fermentation time. However, the content of phosphorus and iron did not show any noticeable change after 72 hrs of fermentation. As fermentation progressed, total sugar and non-reducing sugars decreased (Table 4). The decrease in the pH of *ersho* with fermentation time was not so rapid as they were in the dough. The pH of *ersho* dropped from 3.93 during 48 hrs of fermentation to 3.77 after 96 hrs of fermentation. *Erscho* had a slightly lower pH than that of the tef dough. The fall in pH resulted in increased titratable acidity of *ersho* which rose from 1.83% at 48 hrs fermentation to 2.47% after 96 hrs of fermentation. The volume of *ersho* discarded from the fermenting tef dough significantly ($p<0.05$) increased with the increases in fermentation time (Table 4).

Table 2: Changes in antinutritional factors, minerals, pH and titratable acidity of fermenting tef dough

Time hr	TIA TIU/g	Tannin mg/100g	Phytate mg/100g	Ca mg/100g	P mg/100g	Iron mg/100g	pH mg/100g	% TA
0	5584±67 ^a	881±9 ^a	707±8 ^a	231.5±1.8 ^a	18.4±0.1 ^a	6.1±0.2 ^a	0.14±0.01 ^a	
24	4353±72 ^b	615±4 ^b	510±9 ^b	232.1±2.3 ^a	18.3±0.2 ^a	4.3±0.3 ^b	0.81±0.02 ^b	
48	2761±98 ^c	530±9 ^c	310±5 ^c	214.0±3.7 ^b	15.7±0.1 ^b	4.0±0.1 ^b	0.84±0.03 ^b	
72	2651±83 ^d	471±3 ^d	245±9 ^d	175.6±2.8 ^c	12.5±0.2 ^c	3.8±0.1 ^b	1.05±0.01 ^c	
96	1766±54 ^e	393±8 ^e	198±9 ^e	137.2±1.6 ^d	10.5±0.1 ^d	3.8±0.2 ^b	1.26±0.03 ^d	

Values are means ± S.D. of five determination. Values in each column followed by different superscripts are significantly different ($p<0.05$).

TIA-trypsin inhibitor activity; TA-titratable acidity.

Changes in the titratable acidity of the dough were significantly correlated with NPN, free amino acids and amino acid nitrogen ($p<0.05$).

Discussion

Fermentation of tef dough was spontaneous just as in other traditional cereal dough fermentations during which various biochemical changes take place. The rise in the levels of NPN, free amino acids, soluble proteins and amino nitrogen may be due to hydrolytic action of proteolytic enzymes on dough proteins during tef dough fermentation. The formation of more simple and soluble products after fermentation may enhance available amino acids content, protein digestibility and the nutritional value of tef.

Previous studies also reported that natural fermentation increased the proteolytic activity and the contents of peptides and amino acids resulting in increased protein digestibility and nutritional values of different foods (9, 10, 25). The increase in fat acidity might be due

to increased activity of lipolytic enzymes which produced more free fatty acids which impart their flavour to the final product. A similar observation was reported by Kazanas and Fields (9).

There was a 12% decrease in total protein in fermenting tef dough. The decrease can be attributed to leaching out of the protein into *ersho* which was discarded after 96 hrs of fermentation. Similarly, Gashe et al (3) observed 4 - 13% decrease in total nitrogen content during tef dough fermentation.

Table 3: **Changes in protein components and minerals in *ersho* (mg/ml)**

Time hr	Protein	Non protein nitrogen	amino acid nitrogen	Free amino acids	Ca	P	Iron
48	40.8±0.1 ^a	6.2±0.1 ^a	1.66±0.12 ^a	22.36±0.09 ^a	1.22±0.03 ^a	2.34±0.03 ^a	0.17±0.01 ^a
72	82.4±0.1 ^b	8.4±0.3 ^b	1.96±0.13 ^b	26.17±0.10 ^b	3.44±0.01 ^b	4.00±0.04 ^a	0.34±0.03 ^b
96	120.6±0.3 ^c	14.3±0.2 ^c	2.15±0.10 ^c	29.17±0.07 ^c	4.17±0.02 ^c	3.89±0.01 ^a	0.34±0.02 ^b

Values are means ± S.D. of five determinations. Values in each column followed by different letters are significantly different (p<0.05).

pH and titratable acidity data on fermented tef dough are typical of cereals and legumes undergoing a natural lactic acid fermentation. A rapid drop in pH with a corresponding increase in titratable acidity has been reported in natural fermentation of various food grains (9, 10, 26).

Natural lactic fermentation of tef dough has resulted in a marked decrease in the content of antinutritional factors. The reduction of phytate content of fermented tef dough may be compared with values reported in fermented corn meal (26). Cereal-based diets are usually low in mineral content. The reduction of phytate content during fermentation would make cereal-based diets a good source of iron,

Table 4. **Changes in sugars, pH, titratable acidity and liquid volume in *ersho***

Time hr	Total sugar mg/ml	Redacting sugar mg/ml	Non reducing sugar mg/ml	pH	Titratable acidity %	Liquid volume ml
48	9.05±0.13 ^a	0.41±0.01 ^a	8.74±0.10 ^a	3.93±0.13 ^a	1.83±0.07 ^a	5.32±0.13 ^a
72	7.47±0.11 ^b	.37±0.01 ^b	7.10±0.11 ^b	3.83±0.11 ^a	2.13±0.06 ^b	17.33±0.14 ^b
96	5.71±0.12 ^c	.32±0.02 ^c	5.30±0.14 ^c	3.77±0.12 ^a	2.47±0.11 ^c	23.42±0.13 ^c

Values are means ± S.D. of five determinations. Values in column followed by different letters are significantly different (p<0/05).

zinc and calcium since phytate is known to interfere with the absorption of these minerals.

The diminishing effect on polyphenols of tef dough may be due to the activity of the enzyme polyphenoloxidase elaborated during fermentation which has been reported in *rabadi* fermentation (27). Of the three antinutritional factors, phytic acid appeared to be more susceptible to degradation by fermentation. The extent of reduction, however, in the antinutritional factors appeared to be a function of fermentation time. Decrease in the content of these antinutrients during fermentation may improve the digestibility of proteins, and carbohydrates and enhance the availability of minerals. Traditional fermentation of tef flour, thus, seems to have certain advantages.

Erscho contains protein, amino acids, sugars and minerals. The general decrease in sugar levels in *ersho* with increased fermentation time is possibly due to the metabolism of sugars to glycolytic end products such as lactic and acetic acids. Umata *et al* (28) also reported decreases in starch and sugar contents attributed to the activity of flour amylases during tef dough fermentation. The higher titratable acidity values observed for *ersho* compared to the dough can be attributed to the high buffering capacity of the medium due to the high content of soluble proteins and amino acids. About 41% of calcium, 43% iron and 35% phosphorus in the dough were leached into *ersho* after 96 hrs of fermentation. The concentration of iron in the dough was reduced to 10.5 mg/100 g due to the leaching out of about 8 mg/100 g iron into *ersho*. Consequently, the level of protein, minerals and amino acids in the dough was low. Similarly, minerals and proteins were leached into the steep water during processing of *ogi* (12).

Traditionally, a small portion of *ersho*, which contains a complex group of microorganisms (15), is used as a starter to initiate new fermentation. The use of *ersho* as inoculum may accelerate the fermentation process and is a potential method of improving the availability of nutrients in tef. New approaches, however, need to be sought to fully utilize *ersho*.

This study indicated that certain biochemical changes occur during traditional fermentation of tef flour. Knowledge gained about these changes during the fermentation process is necessary when commercial production of a fermented tef product with constant characteristics and anticipated quality is being considered.

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Development of a tempe-related food using *kocho* and grass pea as substrate

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Abstract: Tempe was prepared by co-fermenting different proportions of *kocho* flour with grass pea using the traditional inoculum, *Usar*. The nutritive quality of tempe prepared from *kocho*-grass pea (20:80; 25:75; 30:70; 40:60; 50:50) was determined. The protein crude fibre and ash content increased significantly, while carbohydrates decreased slightly. The fat content of tempe made from all the *kocho*-grass pea combinations decreased by about one third. Soluble and reducing sugars increased by 2.6- and 2.9-, 4.8- and 9.5- and 7- and 10-fold, respectively, in 30:70, 40:60 and 50:50 *kocho*-grass pea combinations. Free amino acids and non-protein nitrogen similarly increased 8.5- and 24-, 23- and 7.5- and 6.5 and 20-fold, respectively, whereas minerals remained unaffected in 20:80, 25:75 and 30:70 *kocho*-grass pea combinations. Fermentation also significantly decreased the phytic acid and trypsin inhibitors but, increased tannin contents of the tempe. Co-fermentation of *kocho*-grass pea combinations into tempe greatly improved the nutrient of *kocho* in terms of protein, fat and ash contents. The tempe could be used for supplementary feeding. A fermentation scheme was therefore, developed for the production of an enriched product in which 40% to 80% grass pea was fermented with *kocho* flour for 48 hr. [*Ethiop. J. Health Dev.* 1997;11(1):67-73]

Introduction

In developing countries plant proteins are cheaper and more accessible than animal protein and could be used to alleviate protein energy malnutrition (PEM) and other dietetic problems, and improve the low nutritive quality cereals and starch staple foods (1, 2). Grain legumes are rich and cheap sources of dietary protein in developing countries. Grass pea (*Lathyrus sativus* L.) as one of the important legumes contributes more than 7.2% of the total legume production in Ethiopia. Its utilization, however, is undermined by the presence of trypsin inhibitors (3), phytate and tannins causing reduced bioavailability of minerals and prolonged cooking times (4, 5) as well as, β -N-Oxalyl-L- α , β -diaminopropionic acid (ODAP) causing lathyrism (6).

Enset (*Ensete ventricosum*) is one of the most important indigenous crops for Ethiopia. It grows in a wide range of environmental conditions. More than 10 million people in the south, south west and central regions of the country, directly or indirectly, depend on enset for their livelihood (7). *Kocho* is a semi-solid fermented product of enset. It generally contains only 1% or 2% protein and thus is completely unable to provide the consumer with sufficient protein. Being essentially a starch food, *kocho* is not only poor in protein but the protein has also been found to be deficient in the essential amino acids (8). *Kocho* has also another drawback that is, rapid post processing deterioration. The low protein content of *kocho* has been a major concern in its utilization and this has led to *kocho* products being regarded as nutritionally inferior foods. The possibility of increasing the protein content of *kocho* products could help to alleviate this limitation of its wider utilizations.

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Protein quality of starch food may be improved by amino acid complementation with legumes (1,

2) and fermentation (8). *Rhizopus* fermentation of cereal - legume mixture was reported to produce tempe with enhanced nutritive quality (9). Tempe is a popular Indonesian fermented food consisting of tender-cooked soy beans (or occasionally other legumes) bound together in to a white cake by a dense cottony mycelium of the mould *Rhizopus* spp. It owes much of its flavour, sliceable meat-like texture, easy digestibility and excellent nutritional properties to the process of fermentation (10). In Indonesia, tempe has been successfully used in community-based therapy for the management of diarrhoea and rehabilitation of protein energy malnutrition (PEM) cases (11). The synthesis of vitamin B₁₂ (12) during tempe fermentation might also help in anaemia management.

Binyam *et al* reported that the various steps and fermentation procedures used for tempe production from grass pea decreased antinutritional factors significantly including the neucotoxin principle ODAP (5). Tempe is cheap, can be incorporated in many foods, causes no malabsorption syndrome and can be used by all age groups (10).

Investigations have been carried out on methods of improving the protein content of cassava (*Manihot esculanta* Cratz) products by fermentation with protein enriching organisms (13) and legumes (14). Virtually no such study has been carried out on *Kocho*. The aim of the report here was to develop grass pea *Kocho* tempe and estimate its nutrient contents and antinutritional factors.

Methods

Sample preparations: Rice-grown mixed culture *Rhizopus oligosporus* and *Rhizopus oryzae* obtained from the Institute of Sciences, Bandung, Indonesia was used for the inoculation of the substrate.

The local grown grass pea (*Lathyrus sativus* L) was used for this study. The grass pea was dehulled by the traditional grinding mill. *Kocho* purchased from a market in Sidama, Ethiopia, was freeze-dried and ground in a Cyclotec (Tecator, Sweden) sample mill and sieved.

Tempe was prepared according to the procedure developed by Mugula (15) to manufacture sorgum-common bean tempe. Grass pea and *Kocho* were mixed at different weight ratios of *kocho* to grass pea.

A 0.2% inoculum by dry weight of raw materials was used to obtain an acceptable tempe with respect to better mould growth texture and ease of slicing. Incubation was made at room temperature ($28 \pm 2^\circ\text{C}$) in 250 g capacity perforated polyethylene bags. Tempe was sliced into small pieces and freeze-dried for 12 hrs and ground in a Cyclotec sample mill into powder and used for analysis.

Analysis: Proximate composition of samples was analyzed according to AOAC (16) methods. The moisture was estimated by drying to constant weight in the oven at 105°C . The micro-Kjeldahl method was used for analysis of nitrogen and estimation of protein ($6.25 \times \text{N}\%$). Crude fat was extracted with diethyl ether. Ash and crude fiber were determined according to AOAC (16). Carbohydrate was estimated by difference.

Calcium and iron concentrations were determined according to AOAC (16) methods whereas, phosphorus was determined as described by Fiske and Subbarow (17). Phytic acid was extracted with 1.5% HCl in 10% Na₂SO₄ and determined by the method of Haug and Lantsch (18). Tannins were estimated by the modified Vanillin-HCl method of Maxon and Rooney (19) and expressed as catechin equivalent. Trypsin inhibitor activity was determined colorimetrically and expressed as TIU/g dry weight (20). pH of samples was monitored periodically during fermentation using pH meter.

To estimate the soluble carbohydrates, a 10 g sample was suspended in 80 ml deionised water, heated at 70°C in an agitating water bath for one hour and made up to 100 ml. The protein in 20 ml aliquot of homogenate was precipitated with 20 ml saturated lead acetate solution. Soluble carbohydrate supernatants obtained after centrifugation were quantified using the phenol-sulphuric acid method of Dubois *et al* (21) and with glucose standard. Total soluble sugars in the tempe

were extracted with 80% ethanol, then determined using the phenol-sulphuric acid method of Miller (22). Starch was extracted with perchloric acid from the residue and measured according to McCready *et al* (23)

The non-protein nitrogen (NPN) was estimated by the method of Cocon and Soltess (24) and the free amino acids were estimated by the ninhydrin method of Rosen (25).

Statistical analyses: All experiments were conducted three times and analyzed by analysis of variance. Differences were considered statistically significant at $p < 0.05$.

Results

The best tempe was obtained by using grass pea to *Kocho* ratio of 20:80, 30:70, 25:75, 40:60 and 50:50 and 0.2% inoculum. The proximate composition of *Kocho*-grass pea tempe is shown in Table 1. Mould,

fermentation for up to 48 hr apparently increased significantly ($p < 0.05$) the crude protein, fat and ash contents of tempe while it slightly decreased the carbohydrate. Highest increase in protein content was observed in combinations 20:80 (31%) and 25:75 (42%). Fat content in all fermented combinations decreased by about a third compared to the unfermented combinations. The ash content in all fermented mixtures increased significantly ($P < 0.05$) due to fermentation. The total carbohydrate content decreased slightly (4.5-13%) in the fermented mixtures. The crude fibre increased by 31%, 34% and 35% in the fermented 20:80, 30:70 and 50:50 *kocho*-grass pea combinations, respectively. Moisture content of the tempe samples decreased with the increasing proportion of *kocho* in the mixtures. However, fermentation by the tempe mould did not change the content of minerals.

Table 1: Proximate composition of fermented and unfermented *kocho*-grass pea combinations (% dry weight basis)

<i>kocho</i> -grass pea ratio	Process	Moisture	Protein	Fat	Crude fiber	Ash	Carbohydrate
20:80	Unfermented		22.5±1.5 ^a	1.98±0.12 ^a	4.79±0.51 ^a	0.84±0.03 ^a	69.5±2.5 ^a
	Fermented	55.3±2.1 ^a	29.3±1.2 ^b	1.32±0.01 ^b	6.27±0.37 ^b	1.19±0.07 ^b	61.6±3.1 ^b
25:75	Unfermented	-	21.5±1.1 ^c	1.90±0.15 ^a	5.01±0.27 ^c	0.88±0.01 ^a	70.61±1.5 ^a
	Fermented	49.8±2.5 ^b	26.7±2.1 ^d	1.24±0.03 ^c	5.30±0.32 ^c	1.20±0.00 ^b	65.6±1.8 ^c
30:70	Unfermented	-	20.7±1.3 ^e	1.89±0.09 ^a	4.29±0.21 ^c	0.91±0.03 ^a	72.2±5.1 ^d
	Fermented	50.9±2.2 ^c	24.1±1.5 ^f	1.14±0.01 ^d	5.75±0.41 ^e	1.33±0.11 ^b	67.0±1.3 ^e
40:60	Unfermented	-	19.1±0.9 ^g	1.76±0.13 ^e	.49±0.17 ^d	0.98±0.04 ^a	73.4±3.1 ^f
	Fermented	48.2±3.2 ^d	22.4±1.1 ^h	1.11±0.09 ^d	4.69±0.19 ^f	1.40±0.09 ^b	70.4±1.5 ^a
50:50	Unfermented	-	17.4±0.5 ⁱ	1.65±0.14 ^f	3.29±0.12 ^g	0.95±0.05 ^c	76.5±3.2 ^g
	Fermented	46.6±2.3 ^e	20.9±0.9 ^e	1.14±0.10 ^e	.44±0.11 ^d	1.62±0.10 ^d	71.9±1.0 ^h

Values are means of five determinations ±S.D. Values in a column with different letters are significantly different ($p < 0.05$).

The effect of fermentation on carbohydrate components is shown in Table 2. As a result of fermentation, all soluble carbohydrate in all the mixtures increased significantly ($p<0.05$). This was followed by significant decrease in starch content in all the tempe. Highest loss in starch due to fermentation was observed in combinations 40:60 (21.4%) and 50:50 increased 2.6-, 4.8- and 7-fold, respectively. Reducing sugars increased similarly, by about 3- and 10- fold for these combinations. Significant ($p<0.05$) reductions were also observed in available carbohydrate after 48 hr fermentation. Fermentation also resulted in increased pH from an average value of 6.3 to 7.3 in all fermented tempe samples.

As fermentation increased, the quantity of non-protein nitrogen and free amino acids increased significantly ($p<0.05$). Non protein nitrogen increased 24-, 23-, 21-, 17-, and 12- fold, respectively, in all the five *kocho*-grass pea combinations. Similarly, free aminoacids increased 2.4- two 8.5- fold in all the combinations.

Table 2: Carbohydrate components of fermented and unfermented *kocho*-grass pea combinations (% dry weight basis)

<i>kocho</i> -grass pea ratio	Process	Soluble Carbohydrate	starch	Available Carbohydrate	Total Sugars	Reducing Sugars	Nonreducing	pH Sugars
20-80	Unfermented	1.80±0.3 ^a	41.7±0.2 ^a	44.2±1.2 ^a	0.68±0.10 ^a	0.17±0.01 ^a	0.50±0.02 ^a	6.57±0.03 ^a
	Fermented	-	36.8±0.3 ^b	37.5±1.5 ^b	0.82±0.09 ^b	0.29±0.03 ^b	0.53±0.01 ^a	7.23±0.02 ^b
25-75	Unfermented	2.1±0.2 ^b	44.1±0.7 ^a	46.5±2.1 ^a	0.68±0.02 ^a	0.19±0.00 ^a	0.48±0.04 ^b	6.50±0.03 ^a
	Fermented	-	37.6±0.3 ^b	38.6±1.3 ^b	1.24±0.11 ^c	0.31±0.01 ^b	0.93±0.05 ^c	7.20±0.01 ^b
30-70	Unfermented	2.50±0.4 ^c	44.9±0.4 ^a	48.8±2.4 ^d	0.67±0.04 ^a	0.22±0.02 ^c	0.46±0.01 ^b	6.40±0.01 ^a
	Fermented	-	38.7±0.1 ^b	40.3±1.7 ^e	0.77±0.12 ^d	0.63±0.04 ^d	1.14±0.11 ^d	7.03±0.02 ^b
40-60	Unfermented	2.97±0.2 ^d	51.4±0.2 ^c	53.4±3.3 ^f	0.66±0.03 ^a	0.26±0.02 ^e	0.41±0.02 ^e	5.80±0.01 ^c
	Fermented	-	40.4±0.4 ^h	44.0±2.1 ^a	0.85±0.21 ^e	2.48±0.10 ^f	1.37±0.13 ^f	7.17±0.03 ^b
50:50	Unfermented	4.93±0.5 ^e	56.3±0.7 ^e	58.0±2.4 ^g	0.65±0.01 ^a	0.30±0.03 ^g	0.36±0.01 ^g	5.60±0.02 ^c
	Fermented	-	41.8±0.5 ^a	46.4±1.8 ^c	4.54±0.23 ^h	3.03±0.12 ^h	1.51±0.13 ^h	7.23±0.02 ^b

Values are means of five determinations ±S.D. Values in a column with different letters are significantly different ($p<0.05$)

Soluble protein also increased significantly ($p<0.05$) in the tempe samples. Fermentation by the tempe mould also resulted in dry matter losses. The dry matter losses decreased as content of *Kocho* in the combinations increased (Table 4). *Kocho*-grass pea combinations with higher grass pea content have higher trypsin inhibitors. However, fermentation by the tempe mould has decreased significantly ($p<0.05$) this antinutrient in tempe samples. Trypsin inhibitors in 50:50, 40:60 and 30:70 *kocho*-grass pea tempe decreased by 73%, 58% and 56%, respectively.

Fermentation in the fermented *kocho*-grass pea combinations also significantly decreased the phytic acid contents in the 20:80 (80%), 25:75 (79%) and 30:70 (77%) *kocho*-grass pea combinations. However, in fermented 50:50 *kocho*-grass pea combination less (56%) reduction in phytic acid content was observed. Tannin content of the unfermented *kocho*-grass pea combinations appeared to be low, but decreased as the content of grass pea in the unfermented combinations decreased. Fermentation, however, significantly increased tannins in *kocho*-grass pea combinations ($p<0.05$) (Table 4). Such increases in tannin content were evident in the 20:80 (42%), 25:75 (40%) and 30:70 (37%) *kocho*-grass pea combinations.

Discussion

After 17 hr, the *kocho*-grass pea combinations were tied together in rather firm cake by the hyphae. The smell was rather distinctive and may be described as between a yeast-like and a fruit-like aroma. The hyphae seemed even more dense and reached a maximum after 30 hr. After 48 hr, spore formation began and release of free ammonia was noted. Fermentation for 30 to 48 hr, therefore, yielded the most acceptable products.

Fermentation of *kocho*-grass pea combinations by the tempe mould has resulted in various chemical changes. The increase of protein in the present study reflects the decrease of other constituents particularly fat and carbohydrates utilized by the mould. Wang *et al.*(9) attributed the increase in the protein content to the decrease of constituents utilized by moulds for growth and

production of heat during fermentation. The high fiber contents of tempe from *kocho*-grass pea combinations could also be due to enzyme resistant starches. Matsuo (26) also reported an increase in fiber content of *Okara* tempe manufactured using *R. oligosporus*.

During prolonged fermentation (0 to 48 hr), starch decreased significantly ($p<0.05$) in all combinations of the *Kocho*-grass pea tempe

Table 3: Nintrogenous constituents, soluble protein and dry matter loss of fermented and unfermented *kocho*-grass pea combinations (dry weight basis)

<i>kocho</i> grass pea ratio	Process	NPN%	Free amino acids mg/g	Soluble protein %	Soluble solids %	Dry matter loss%
20:80	Unfermented	0.10±0.01 ^a	11.2±1.5 ^a	1.88±0.02 ^a	7.14±0.10 ^a	-
	Fermented	2.40±0.11 ^b	93.3±1.4 ^b	19.63±0.63 ^b	26.03±4.1 ^b	12.0±2.1 ^a
25:75	Unfermented	0.10±0.01 ^a	11.6±1.1 ^a	1.83±0.05 ^a	6.91±0.11 ^a	-
	Fermented	2.26±0.13 ^b	87.2±3.5 ^c	17.43±0.41 ^c	25.90±3.2 ^b	10.7±1.7 ^b
30:70	Unfermented	0.09±0.01 ^a	12.3±0.9 ^d	1.77±0.04 ^d	6.66±0.12 ^c	-
	Fermented	1.91±0.02 ^c	79.3±5.3 ^e	14.50±0.39 ^e	23.40±1.3 ^d	8.8±2.3 ^c
40:60	Unfermented	0.09±0.02 ^a	13.6±0.7 ^f	1.66±0.03 ^f	6.18±0.10 ^e	-
	Fermented	1.55±0.04 ^d	56.4±5.3 ^g	8.17±0.12 ^g	21.77±1.2 ^f	5.7±1.1 ^d
50:50	Unfermented	0.08±0.01 ^a	14.8±0.8 ^h	1.55±0.01 ^h	5.70±0.12 ^g	-
	Fermented	1.40±0.02 ^d	34.8±5.6 ^f	6.83±0.20 ^f	20.00±1.5 ^h	3.6±0.9 ^e

Values are means of five determinations ±S.D. Values in a column with different letters are significantly different ($p<0.05$).

with concomitant significant ($p<0.05$) increases in total and reducing sugars. These changes may be attributed to the amylolytic activity of the mould which increased the content of sugars by starch hydrolysis and its fermentation to organic acids (27). Berghofer (28) also reported significant decrease in starch during field bean tempe preparation.

The tempe mould also possesses a strong lipolytic activity hydrolysing fermentation of *kocho*-grass pea combinations. The lipolytic activity of tempe mould was also reported by Wagenkeench et al. (29). The pH showed a progressive increase to more than 7.0, which could be largely attributed to the active proteolysis and deamination of amino acids by the mould. The increase in pH with fermentation time is not unusual, since a similar trend occurs in tempe fermentation (29).

The proteolytic activity of tempe mould could also account for the significant ($p<0.05$) increases in free amino acids and non-protein nitrogen content of fermented *kocho*-grass pea combinations. Matsuo (26) reported an increase in soluble nitrogen/total nitrogen ratio, free fatty acids/crude fat ratio, and free sugar carbohydrate ratio on frementation of *okara* into tempe.

Because of enzymatic degradation of macromolecules into substances of lower molecular weight, there is solubilisation effect in the *kocho*-grass pea tempe samples. Due to the digestion of the martix between the *kocho* and grass pea cells, a significant reduction in total dry matter and increases in total solube solids, soluble protein and solube carbohydrate were observed. Similar modifications took place in common beans (30), chickpeas and horse beans (31) when made into tempe.

Fermentation for 48 hr significantly ($p<0.05$) decreased the antinutritional factors, trypsin inhibitors and tannins, in the tempe samples. The significant loss in trypsin inhibitor contents in fermented *kocho*-grass pea combinations may be attributed to the capability of the tempe mould to hydrolyse the trypsin inhibitors of the combinations. Wang et al. (32) also reported that *R. oligosporus* is capable of hydrolysing the trypsin inhibitors of soy beans. Tannin content of unfermented *kocho*-grass pea combinations appeared to be low. However, fermentation for 48 hr caused a significant ($p<0.05$) increase in the content of this antinutrient. The increase in tannin content of the fermented combinations may be due to the activity of tempe mould enzymes to relaease tannins from tannin-protein complexes during fermentation. Mugula (15) also reported a 42% increase in tannins during the fermentation of sorghum-common bean mixtures into tempe. From the nutritional point of view, tannins depress growth rate, lower protein digestibility, amino acid availability and increase faecal nitrogen (33).

Fermentation also caused significant ($p<0.05$) reduction of phytic acid in tempe

Table 4: Minerals and antinutritional factors in unfermented and fermented *kocho*-grass pea combinations (dry weight basis)

<i>kocho</i>	Process	Fe mg/100g	Ca mg/100g	P mg/100g	Tannins	Phytic acid	Trypsin
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grass pea ratio					Mg/100g	mg/100g	inhibitors IU/g
20:80	Unfermented	2.6±0.3 ^a	131±3 ^a	351±3 ^a	124±3 ^a	880±51 ^a	8088±113 ^a
	Fermented	2.6±0.2 ^a	130±2 ^a	357±3 ^a	176±6 ^b	173±19 ^b	4285±131 ^b
25:75	Unfermented	2.6±0.5	31±5 ^a	346±5 ^a	120±3 ^c	825±38 ^c	7967±223 ^a
	Fermented	2.6±0.4 ^a	126±2 ^a	341±5 ^a	168±4 ^d	174±17 ^b	3957±103 ^c
30:70	Unfermented	2.7±0.6 ^a	131±3 ^a	342±4 ^a	115±2 ^e	770±42 ^d	7697±317 ^d
	Fermented	2.6±0.1 ^a	134±4 ^a	349±0 ^a	158±5 ^f	80±9 ^b	3404±97 ^e
40:60	Unfermented	2.7±0.7 ^a	131±5 ^a	353±6 ^a	105±3 ^g	660±34 ^e	7158±231 ^f
	Fermented	2.6±0.3 ^a	133±3 ^a	345±1 ^a	138±9 ^h	392±21 ^f	2993±177 ^g
50:50	Unfermented	2.6±0.4 ^a	131±2 ^a	334±7 ^a	96±6 ^f	550±23 ^g	6619±212 ^h
	Fermented	2.6±0.6 ^a	133±6 ^a	347±4 ^a	125±3 ^g	569±27 ^g	1791±87 ^f

Values are means of five determinations ±S.D. Values in a column with different letters are significantly different (p<0.05)

from *kocho*-grass pea combinations which may be due to endogenous and mould phytase. Production of phytase by the fungus is well documented (11). The decrease in phytate content would increase the bioavailability of minerals particularly zinc and iron which might be important in the management of anaemia.

The present study has shown that high potential exists for the use of tempe fermentation in improving the nutritional quality of starch foods. The co-fermentation of *kocho* with grass pea in the present study has dual advantages in that protein content was increased while the antinutritional factors content of the *kocho*-grass pea combination decreased, resulting in more nutritive and safer food product. The use of such improved product is suggested at a household level as a way to improve the nutritive value of enset foods such as *kocho* and *bulla*.

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Natural fermentation of Enset (*Ensete ventricosum*) for the production of Kocho

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Abstract: Biochemical changes of fermenting enset were studied. After seven weeks fermentation, total protein, ash and total carbohydrates decreased by 15%, 16% and 34%, respectively. Significant ($p<0.05$) reduction in iron (15%), phosphorus (29%), calcium (51%), starch (23%), soluble sugars (93%), reducing-sugars (84%) and available carbohydrates (51%) were recorded. Free amino acids and non-protein nitrogen increased by 6- and 1.6-fold, respectively. The pH of the fermented mash fell from an initial value of 5.7 to 3.8 with a concomitant sharp rise in titratable acidity resulting in accumulation of organic acids. The predominant organic acids were lactic, iso-valeric and n-butyric acids followed by n-valeric and acetic acids. Other volatile fatty acids and ethanol were formed in lesser quantities. Fermentation of enset also resulted in significant reductions in tannins and trypsin inhibitors whereas oxalic acid remained unaffected. [*Ethiop. J. Health Dev.* 1997;11(1):75-81]

Introduction

In Ethiopia there are several traditionally fermented foods some of which are products of a low-technology process involving a solid state fermentation (SSF) system. The technique is usually cheap and simple to operate but can be time-consuming and labour-intensive. Biochemically, SSF is a complex system that has not been fully characterized and understood. The technology has been used in Ethiopia to produce such food items as kocho and bulla.

Kocho and bulla are naturally fermented food products of enset (*Ensete ventricosum*). Enset is a perennial root crop which grows between altitudes of 1500-3000 meters above sea level (1). It provides an essential source of calories for as many as 10 million people in the South and Southwestern parts of Ethiopia (2, 3).

One of the processing methods of enset involves the fermentation of the corm and pseudo stem in an underground pit for several weeks during which period they soften and turn into a whitish, semi-solid dough-like

meal popularly called *Wassa* or *Kocho*. This dough-like meal (product) is prepared into various types of foods. Selinus *et al* (1) have listed food types prepared from *kocho*. *Kocho* is consumed widely by people of all social classes in South and Southwestern Ethiopia. Fermentation of enset is a microbial disintegration of the grated corm and pseudo-stem and is reported to impart flavour and textural qualities to the fermented food products, *kocho* and *bulla*. The odour change is much more pronounced in this fermentation than any other fermented food products locally encountered. Studies made to date on fermented enset products of *kocho* and *bulla* have tended to concentrate on carbohydrate and calcium composition (4) amino acid pattern (5) and identification of the microflora involved in enset fermentation processes (6, 7, 8). Virtually nothing is known about the biochemical changes due to fermentation and the content

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of antinutritional factors in fermented enset. This paper reports the biochemical changes brought about by natural lactic fermentation of enset into *kocho* including the amounts and types of organic acids produced during fermentation.

Methods

Enset fermentation: Enset plants of variety Ado were purchased from a farmer in Sidama. All the fermentation processes were carried out in the enset farm at the backyard of the farmer. Enset was fermented using the traditional *wassa* fermentation procedure of Sidama as described by Selinus et al (1). This involved scrapping of the leaf bases with sharp-edged bamboo split (*Sissicho*) to extract the long fibres. The liquid oozing out was collected in a separate pit lined with fresh enset leaves. Water was decanted after three days and the starch residue (*bull*) wrapped with enset leaves and kept in the fermentation pit with “*Amulcho*” to ferment. Part of the enset corm pulverized with rough-edged serrated wooden piece (*Keho*) was mixed with the leaf scrapping locally called *Amulcho* or *Ambicho*, covered with enset leaves and stored in the open air for five to seven days. The remaining part of the corm is rubbed with decomposed enset leaves (*Shigido*), recovered from the enset farm, covered with fresh leaves and left for four to five days in the open air. The retted corm is pulverized into fine pieces and thoroughly mixed with *Amulcho*. The mixture is then transferred to a fermentation pit lined with layers of fresh enset leaves, covered with same and left to ferment. The first sample, referred to as zero week (Ow) sample, was collected immediately after the mash was placed in the pit, thereafter samples were obtained at intervals of one week.

Chemical analyses: To measure the pH, a 10 g portion of the fermenting enset mash was homogenized with deionized water and the suspension measured using a pH meter (Orion, USA) equipped with a glass electrode.

The titratable acidity (TA) expressed as percent lactic acid was determined by titrating 25 g of the decanted homogenate samples used for pH determination against 0.1 N NaOH to pH 8.3.

Alcohols were determined by gas liquid chromatography on a Varian 3700 VISTA (Varian Associates, USA) instrument equipped with a flame ionization detector using a glass column (2mx2mm, i.d.) packed with Propack QS and temperature programmed from 120°C to 220°C at 20 min⁻¹ with nitrogen as carrier gas at a flow rate of 40 ml min⁻¹. Pentan-2-ol was used as an internal standard and quantisation was performed with the aid of a Varian 4270 computing integrator.

Volatile fatty acids were determined by gas liquid chromatographic analyses of the aqueous extracts of fermenting enset according to Rogossa and Love (9) and Patrick and Timothy (10). A column (2m x 3.2mm i.d.) of 15% FFAP on Chromosorb WAW - mesh 80/100 held at 180°C and isothermal operation was used. Injector port, and detector temperatures were 210°C and 220°C, respectively. 2-Methyl valeric acid was employed as internal standard and quantisation was performed as before. Lactic acid was estimated by the method of Alcock (11).

Table 1: Effect of fermentation on proximate composition of *kocho* (dry weight basis)

Period	Moisture	Total protein	Fat	Crude fiber	Ash	NPN	Free amino acids
wk	%	%	%	%	%	%	mg/g
0	84±3	4.07±0.02	0.43±0.01	3.43±0.03	0.75±0.01	0.01±0.01	8.1±1.2
1	66±4	3.97±0.01	0.43±0.00	4.07±0.02	0.76±0.03	0.01±0.00	9.0±1.0
2	63±2	3.87±0.03	0.42±0.00	4.13±0.03	0.78±0.04	0.03±0.00	10.7±0.8
3	60±5	3.75±0.01	0.41±0.01	4.21±0.05	0.75±0.03	0.05±0.02	12.4±0.9
4	60±3	3.63±0.00	0.53±0.04	4.33±0.02	0.67±0.00	0.06±0.01	16.9±1.0
5	60±7	3.53±0.04	0.43±0.05	4.37±0.03	0.65±0.02	0.06±0.00	18.8±1.2
6	60±2	3.47±0.03	0.43±0.02	4.50±0.05	0.64±0.01	0.07±0.01	20.0±0.8
7	60±4	3.47±0.01	0.43±0.05	5.00±0.04	0.63±0.04	0.07±0.01	20.7±0.7

Values are means±S.D. of three determinations.

For analysis of proximate composition, the fermenting enset mashes were freeze dried and ground in a Cyclotec (Tecator, Sweden) sample mill and kept in glass jars at 4°C until used for analysis.

The samples were analyzed for proximate compositions by AOAC (12) methods. Crude protein was calculated using the factor 6.25. Carbohydrate content was determined by difference.

Total sugars other than starch were extracted in ethanol by reflux method and were estimated colorimetrically according to Dubois *et al* (13). Starch from sugar free pellet was estimated by the method of McCready *et al* (14). Reducing-sugars were estimated by the method of Miller (15) and non-reducing- sugars calculated by the difference between total sugars and reducing sugars.

The method of Vanillin-HCl described by Maxon and Rooney (16) was employed for the examination of tannin content using catechin as a standard and expressed as catechin equivalents (mg/100 g, dry wt).

The trypsin inhibitor activity was determined by the method of Kakade *et al* (17) using N-benzoyl-DL-arginine-p-nitroaniline (BAPNA) as the trypsin substrate and expressed as TIU/g (dry wt). Oxalic acid in the fermenting onset was determined by precipitation with calcium salt and potassium permanganate titration (12).

Total iron was determined using bathophenanthroline as described in AOAC (12), phosphorus by the Fiske-Subbaraw method (18) and calcium according to the AOAC (12).

The ninhydrin method of Rosen (19) was used to estimate free amino acids with a standard of leucine. Non-protein nitrogen (NPN) was determined by the procedure of Concon and Soltess (20).

Statistical analysis: Tests were replicated three times. Data were subjected to analysis of variance (ANOVA). Differences were considered statistically significant at $p < 0.05$.

Table 2: Effect of fermentation on carbohydrate components of kocho (% dry weight)

Period	Starch	Total Sugar	Reducing sugar	Non-Reducing Sugar	Available carbohydrate
0	68.8±2.5	4.3±0.1	1.9±0.3	2.4±0.4	72.1±0.4
1	68.5±1.4	4.0±0.4	1.5±0.1	2.5±0.3	72.1±0.5
2	65.9±3.7	3.5±0.3	1.2±0.2	2.3±0.0	72.0±0.7
3	57.3±5.1	6.2±0.7	2.6±0.4	3.6±0.2	61.5±0.6
4	55.3±0.7	4.5±0.8	0.8±0.0	3.7±0.4	57.8±0.8
5	53.3±1.2	1.2±0.1	0.5±0.0	0.7±0.1	54.1±0.5
6	53.1±0.5	1.1±0.2	0.3±0.1	0.8±0.2	54.0±0.9
7	53.1±1.2	0.3±0.0	0.3±0.0	--	54.0±0.7

Values are means±S.D. of three determinations

Results

The fermentation of onset is one of the most important steps in kocho preparation. *Amulcho* has no smell or characteristic flavour when transferred to the fermentation pit. When the pH of the mash reached 4.4 with 0.96% titratable acidity (as lactic acid), the desired sour flavour and characteristic aroma was attained. The proximate composition of fermenting onset are shown in Table 1. Fermenting onset as it was initially prepared (week 0) had a high moisture content (84%) and a near neutral pH (6.5). However, at week seven, when the fermentation reached the desired fermentation stage, the moisture content significantly ($p < 0.05$) decreased to about 60%. The fermentation process also resulted in a significant ($p < 0.05$) losses of protein, ash, and total carbohydrate the losses being 15, 16, and 34%, respectively, after seven weeks of fermentation. Fat content remained unaffected whereas crude fibre increased by 30%. The loss in ash content of the fermenting onset similarly resulted in significant ($p < 0.05$) losses of iron (51%), phosphorus (29%) and calcium (51%), respectively.

As fermentation progressed, the free amino acids and non-protein nitrogen increased to values which were six times and 1.6 times, respectively, the sample at week zero. A significant ($p < 0.05$) and positive correlation was observed regarding the relative levels of free amino acids and the amounts of non protein nitrogen in *kocho*.

The changes in carbohydrate components are shown in Table 2. The starch content significantly ($p < 0.05$) decreased from a starting value of 69% to 53% at the end of the fermentation. During fermentation, sugars were not completely depleted. Two distinct patterns of

sugars consumption can be observed. In the first case, there was an increase in sugars in the first three weeks of fermentation. In the second case, the accumulation of sugars is not observed. After seven weeks of fermentation period, the concentrations of total and reducing sugars declined to about 0.3%. The changes in starch and sugars resulted in a significant loss of available carbohydrate (51%) ($p<0.05$).

Fermentation also resulted in a sharp increase in titratable acidity from the first week to the 6th week of fermentation and it

Table 3: **Production of organic acids during onset fermentation (mg/100g dry weight basis)**

Period wk	Acetic acid	Propionic acid	n-Butyric acid	i-valeric acid	n-valeric acid	-caproic acid	i Lactic acid	Ethanol	TA %	pH
0	8.5±1.5	3.8±0.2	5.0±0.3	8.7±0.1	-	-	51.3±2.1	5.3±0.3	0.27±0.02	5.7±0.1
1	1.0±1.7	4.5±0.3	8.1±0.2	23.1±0.4	-	-	126.4±3.1	5.4±0.1	0.30±0.01	5.1±0.1
2	3.8±1.9	5.3±0.2	10.5±0.5	25.4±0.8	2.5±0.8	-	67.8±2.7	5.7±0.3	0.52±0.83	4.9±0.2
3	6.0±2.0	6.6±0.3	27.5±1.2	60.0±2.1	8.9±0.3	-	90.2±3.7	6.3±0.2	0.96±0.04	4.4±0.2
4	9.1±2.1	8.9±0.4	34.4±1.3	63.7±3.2	15.5±0.4	-	210.0±3.1	12.1±0.4	0.98±0.06	4.2±0.1
5	3.5±2.6	9.5±0.6	35.3±1.2	65.5±3.0	22.0±0.4	1.7±0.2	486.1±4.3	15.0±0.2	1.28±0.01	4.0±0.1
6	1.4±2.1	10.3±0.7	35.6±1.2	66.9±2.1	24.4±0.5	2.6±0.1	504.0±3.5	15.3±0.5	1.50±0.20	3.9±0.2
7	1.9±2.0	12.5±0.6	35.7±2.0	68.9±1.8	26.3±0.6	3.0±0.4	520.4±3.0	21.0±0.4	1.54±0.40	3.8±0.1

Values are means of five determinations ± S.D.

gradually levelled off at the 7th week (Table 3). At the same time, pH also decreased in a similar manner broadly paralleling total acid accumulation in the fermentation mixture. A significant negative correlation was observed between pH and titratable acidity ($p<0.05$).

The major organic acids produced were acetic, lactic, n-butyric, propionic, iso-valeric, and iso-caproic acids. Ethanol was the only alcohol detected during fermentation but the compound attained a maximum concentration of only 21mg/100 g (dry weight basis) after seven weeks of fermentation rendering it unlikely that yeasts are involved in anaerobic degradation of substrate. Lactic and iso-valeric acids were the predominant organic acids and accounted for more than 84% of the total organic acids followed by n-butyric and acetic acids. The concentrations of n-valeric, iso-hexanoic and propionic acids appeared to be low.

Table 4 shows that the content of tannins and trypsin inhibitor activity in fermenting onset was decreased significantly ($p<0.05$). The reduction was about 51% for tannins and 53% for trypsin inhibitor activity at the end of the fermentation period. However, the content of oxalic acid remained unaffected during fermentation of onset.

Discussion

Fermentation of onset into *kocho* is one of the most important processing steps in onset utilization. During the onset fermentation, significant losses in protein, ash and carbohydrates were observed. Most of the losses may be attributed to the leaching of these nutrients through the permeable fermentation pit. Abraham *et al* (5) and Taye (4) also observed losses in protein and minerals during the fermentation of onset.

An increase in the concentrations of nonprotein nitrogen and free amino acids was also apparent as a result of fermentation, a finding which duplicates observations during the *ogiri* fermentation (21). Such significant increases may be accounted for by the activity of proteolytic fermenting onset.

During the onset fermentation process, a progressive decline in carbohydrate content was observed. This may be due to the activity of the microflora which most likely derived its energy from carbohydrate metabolism. The level of soluble and reducing sugars increased by about 1.4-fold during the first three weeks of fermentation and then decreased significantly at the end of the fermentation period. The increase in sugars content in the first case is probably due

to increased activity of native or microbial amylases which hydrolysed starch into sugars (22). The decrease in sugar levels thereafter is probably due to the metabolism of these sugars to glycolytic end products such as lactic acid and other volatile organic acids. Similar trends were observed during the fermentation of cassava (23, 24).

Table 4: **Effect of fermentation on minerals and antinutritional factors of kocho (dry weight basis)**

Period	Ca mg/100g	Iron mg/100g	P mg/100g	Tannin mg/100g	TIA TIU/g	Oxalic acid mg/ 100g
0	288.0±23	3.4±0.1	154.3±11	676±41	8356±11	230±32
1	283.7±23	2.8±0.3	149.0±10	644±34	8137±120	228±23
2	271.7±17	2.5±0.4	140.7±12	440±23	7954±177	225±24
3	210.0±13	2.4±0.0	133.3±9	430±31	6520±123	224±22
4	186.3±21	2.4±0.0	131.7±11	422±30	5184±130	220±21
5	157.3±20	2.2±0.3	131.3±7	390±32	4288±137	224±19

Enset fermentation into *kocho*, like cassava, is primarily a lactic fermentation (25). This is indicated by the sharp drop in pH and rapid rise in titratable acidity. The fall of pH into the acid range could be attributed to an increased amylolytic activity of enzymes causing liberation of acids and possibly other acidic end products of carbohydrates fermentation (26, 27). Amund *et al* (22) similarly reported that the available sugars were transformed into acid during the natural fermentation of cassava.

Fermented enset, *kocho*, has a characteristic penetrating butyrous smell. Although these minor acids contribute only a small amount to the total organic acids, they make significant contribution to the flavour of the *kocho* because of their characteristic odours.

Various parallels may also be drawn between lactic acid fermentation of cassava and enset. In cassava fermentation, lactic and acetic acids comprise most of the total organic acids while other minor volatile fatty acids (propionic, iso- and n-butyric acids) contribute less (25). However, the presence of iso-valeric and n-butyric acids in fermenting enset which may impart the peculiar aroma to *kocho*, in appreciable amounts is a difference.

The bases and corms of enset show enzymic browning reactions when cut and exposed to the air. This can occur with the corm in the fresh state, after storage or when physiologically damaged which can be associated with the reactions of phenolic substances. The diminishing effect on tannins during enset fermentation may be due to the activity of polyphenoloxidase enzyme in fermenting enset. Dhankher and Chauhan (28) also reported decreases in the polyphenols of *rabadi* with an increase in fermentation time.

Trypsin inhibitor activity was significantly ($p < 0.05$) reduced due to fermentation of enset. On the other hand studies on the stability of trypsin inhibitors to cooking or baking of *kocho* is required which may throw light on the problem. The presence of appreciable levels of oxalic acid, trypsin inhibitors and tannins in enset that contributes the major component of the diet of the South and South Western people of Ethiopia is likely to limit its utilization. However, natural fermentation of enset markedly reduced the content of trypsin inhibitors and tannins thus enhances the digestibility of *kocho* protein and bioavailability of minerals. This study has provided useful data. However, there is a need for greater understanding on changes brought about by natural fermentation of enset as the trend in this fermentation is to devise ways to shorten *kocho* production time.

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Original article

Microbial load and microflora of "chat" (*Cata edulis* Forsk) and effect of "chat" juice on some food-borne pathogens

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Abstract: Chewable leaves of "chat" (*Cata edulis*) were collected from retail shops in Awassa and analyzed for their microbial load and microflora in March - June, 1995. Aerobic mesophilic bacteria, Enterobacteriaceae and bacterial spores had mean counts of >10⁴ cfu/g. *Staphylococcus* sp., *Bacillus* sp. and yeasts and molds had mean counts of 10³ cfu/g. Between 60% and 80% of the "chat" samples yielded Enterobacteriaceae, *Bacillus* sp., *Micrococcus* sp. and *Staphylococcus* sp. The aerobic mesophilic flora was dominated by Enterobacteriaceae (25%), *Bacillus* sp. (22%), *Micrococcus* sp. (18%) and *Staphylococcus* sp. (13%). About 47% of the *Staphylococcus* isolates were *Staphylococcus aureus*. "Chat" juice retarded growth of *Salmonella typhimurium*, *Salmonella enteritidis* and *Staphylococcus aureus* at higher concentrations, but complete inhibition was not attained even at 100% concentration. *Bacillus cereus* was inhibited at lower concentrations (25%). Inhibition of *Listeria monocytogenes* was observed only at 100% concentrations. [Ethiop. J. Health Dev. 1997;11(1):83-87]

Introduction

"Chat" (*Cata edulis*) is an evergreen shrub of the family Celastraceae that grows primarily in

Ethiopia, Kenya and Yemen. In the literature, "chat" is consistently referred to as khat and is socially and economically one of the most important plants not only of many countries of Eastern and Southern Africa but also of the Middle East (1). Only the fresh leaves have the desired effect and the "chat" habit has, thus, remained endemic to these areas (2). Due to the development of international air travel, "chat" use has spread to countries far away from the areas of cultivation (2). The juices of the fresh leaves are ingested and this produces central stimulation in man. Between five and ten million people are reported to chew "chat" (2).

In Ethiopia and neighboring countries, "chat" is commonly consumed for social recreation (3) and for other purposes by various occupational groups and students (4). It is also chewed for religious and medicinal uses (5).

There are various studies on the chemical and pharmacological nature of "chat" (2,6). Studies on clinical effects of "chat" have shown that "chat" chewing results in significant increase in blood pressure and the medical and psychological effects are hazardous both to the individual and the community (7).

In Ethiopia, "chat" is consumed without any pre-treatment. Considering the non-hygienic transportation and wrapping processes of this plant from farm-hand to distributors, whole sellers, retailers and finally the consumer, "chat" is exposed to various sources of contamination. It may, thus, be contaminated by a variety of microorganisms including those that can cause food-borne

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diseases. The purpose of this study was, therefore, to determine the microbial load and microflora of "chat" and evaluate if the "chat" juice could eliminate some important food-borne pathogens.

Methods

Collection of Samples: A total of 51 fresh "chat" samples were collected in sterile plastic bags from different "chat" retail markets in Awassa, Ethiopia. The samples were immediately brought to the laboratory and processed for the following microbiological parameters.

Microbiological Analyses: Ten grams of fresh chat leaves were mixed with 90 ml of sterile deionized water in a sterile screw cap bottle and shaken manually for over two minutes to obtain a homogenized washed sample suspensions.

Aerobic mesophilic bacteria: Samples were further diluted in sterile water and volumes of 0.1 ml of appropriate dilutions were spread-plated in duplicate on pre-dried surfaces of Plate Count Agar (PC; Merck) with a bent glass rod. Colonies were counted after incubation at 30 to 32°C for 48 h.

Enterobacteriaceae: Volume of 0.1 ml of appropriate dilutions were spread plated in duplicate on pre-dried surfaces of Violet Red Bile Glucose Agar (Oxoid) plates. The plates were incubated at 30 to 32°C for 24 h.

Bacillus cereus: Volumes of 0.1 ml of appropriate dilutions were spread plated in duplicates on pre-dried surfaces of *Bacillus cereus* agar (Oxoid) and colonies were counted after incubation at 30 to 32°C for 24 h.

Staphylococci: Appropriate dilutions were spread-plated on duplicate plates of Mannitol Salt Agar (Oxoid) and incubated at 30 to 32°C for 48-72 h. Ten colonies from countable plates were picked and slide and tube coagulase test was done to identify *Staphylococcus aureus*.

Bacterial spores: Tubes containing 10 ml of suspensions were heat shocked in a water bath at 80°C for 15 min. A volume of 0.1 ml of the pasteurized sub-samples was streak plated on pre-dried surfaces of Plate Count Agar and incubated at 30-32°C for 48 h.

Yeasts and molds: Volume of 0.1 ml of appropriate dilutions were spread-plated in duplicate on pre-dried surfaces of Chloramphenicol-Bromophenol-Blue agar (CBB) consisting of (g/l in distilled water): yeast extract, 5.0; glucose, 20.0; chloramphenicol, 0.1; Bromophenol Blue, 0.01; agar, 15; pH, 6.0 to 6.4. Yeast colonies were counted after incubating the plates at 25-27°C for 5

d.

Flora assessment: After colony counting, 10 to 15 colonies were selected at random from countable PC agar plates. The sub-cultures were further purified by repeated plating. A total of 532 strains were isolated and tentatively differentiated into various bacterial groups by the following characteristics: phase-contrast microscopy was used to examine cell shape and grouping, presence or absence of endospores and motility; Gram reaction was determined using the KOH test of Gregersen (8); cytochrome oxidase was tested by the method of Kovacs (9); catalase test was made with 3% (v/v) H₂O₂ solution; and glucose metabolism was investigated by the O/F test of Hugh & Leifson (10).

Preparation of "Chat" Juice: Juice from fresh consumable "chat" leaves was extracted by fruit grinding centrifugal machine and steam-sterilized. As preliminary observations had indicated that filter-sterilized and steam-sterilized juice had no noticeable difference in activity on test strains, steam sterilized juice was used in subsequent experiments. The juice was diluted in Brain Heart Infusion (BHI) Broth (Merck) to give final concentrations of 25%, 50% and 75% "chat" juice. Undiluted juice was 100% and BHI broth without "chat" juice served as a control.

Table 1: Microbial load (log cfu/g) of fresh chewable "chat" leaves.

Bacterial groups	$\bar{x} \pm \text{S.D.}$	%C.V.
AMB	5.86 ± 0.82	14
Bacterial spores	4.79 ± 0.91	19
Enterobacteriaceae	4.04 ± 1.25	31
<i>Staphylococcus</i> sp.	3.35 ± 0.84	25
<i>Bacillus cereus</i>	2.19 ± 0.22	10
Yeasts and molds	3.19 ± 0.61	19

AMB, Aerobic mesophilic bacteria

$\bar{x}$, Mean

S.D., Standard deviation

C.V., Coefficient of variation

The pH of the juice was measured by dipping an electrode of a pH meter into the juice.

Test Organisms: The following bacterial test strains were used in this study. *Salmonella typhimurium* (A 13), *Salmonella enteritidis* (A 2), *Staphylococcus aureus* (WS 1759), *Bacillus cereus* (WS 1537) and *Listeria monocytogenes* (WS 2300). The cultures were obtained from the culture collections of Bakteriologisches Institut, SVFA, Weihenstephan, Federal Republic of Germany.

Table 2: Frequency distribution of dominant aerobic mesophilic bacteria from fresh chewable "chat".

Bacterial groups	Positive samples		Isolates	
	No.	%	No.	%
Enterobacteriaceae	41	80.4	133	25.0
<i>Bacillus</i> sp.	35	68.6	115	21.6
<i>Micrococcus</i> sp.	38	74.5	97	18.2
<i>Staphylococcus</i> sp.	32	62.7	68	12.8
Coryneforms	51.0	37	7.0	
<i>Aeromonas</i> sp.	19	37.3	20	3.8
<i>Pseudomonas</i> sp.	12	23.5	18	3.4
<i>Streptococcus</i> sp.	8	15.7	16	3.0
<i>Lactobacillus</i> sp.	10	19.6	15	2.8
<i>Acinetobacter</i> sp.	8	15.7	13	2.4

Inoculation of "chat" juice with test strains: Overnight cultures of the test strains were separately inoculated in the various concentrations of "chat" juice and the control tubes to give a final inoculum level of 102 - 103 cfu/ml. The inoculated tubes were mixed thoroughly and incubated at 30-32°C for 8 h.

Analyses of samples: Appropriate dilutions from freshly inoculated tubes were surface plated on Brain Heart Infusion agar (MERCK) in duplicate to determine the initial inoculum level. The various tubes were then sampled after four and eight hours. Inoculated tubes were sampled (1 ml) aseptically and appropriate dilutions were spread-plated on the following media (all from OXOID). XLD agar for *Salmonella*, Mannitol Salt agar for *Staph. aureus*, Modified McBride agar for *L. monocytogenes* and *Bacillus cereus* agar for *B. cereus*. Inoculated plates were incubated at 32°C for 24-48h for colony counting.

Results

Of the 51 "chat" samples, 29 had aerobic mesophilic counts of $>1.0 \times 10^6$ cfu/g. The mean count was 7.2×10^5 cfu/g with little variation in counts among samples (C.V., 14%). Enterobacteriaceae and bacterial spores had mean counts of around 104 cfu/g and *Staphylococcus* sp., *Bacillus cereus*, and yeasts and molds had counts of 103 cfu/g. Marked variations were noted in counts of Enterobacteriaceae and *Staphylococcus* sp. among samples (C.V., 25%) (Table 1).

The aerobic mesophilic flora consisted of a variety of microorganisms (Table 2) and the dominant ones were Enterobacteriaceae (25%), *Bacillus* sp. (22%), *Micrococcus* sp. (18%) and *Staphylococcus* sp. (13%). Enterobacteriaceae, *Micrococcus* sp., *Bacillus* sp. and *Staphylococcus* sp. were isolated from 80%, 75%, 69% and 63% of the "chat" samples, respectively.

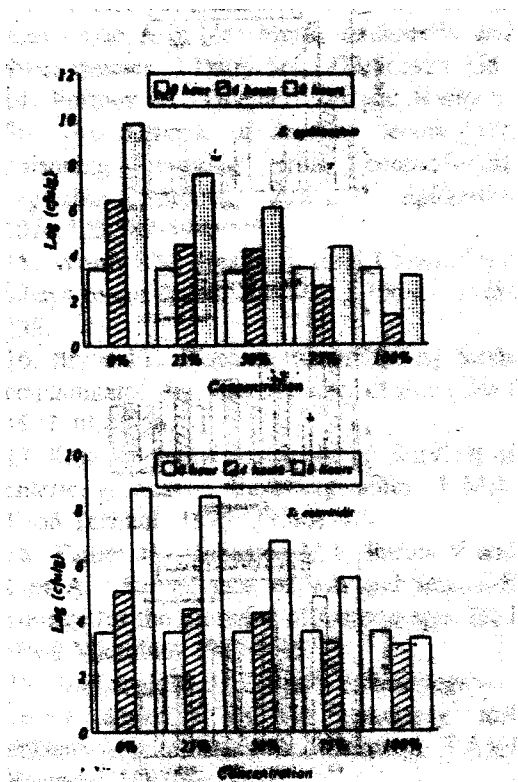


Figure 1: Fate of *S. typhimurium* and *S. enteritidis* at different concentrations of "chat" juice.

About 47% of the *Staphylococcus* isolates were found to be *Staphylococcus aureus* and the remaining were coagulase negative staphylococci.

"Chat" juice had pH value of 6.2. All test strains grew luxuriously in media containing no "chat" juice. Retardation of our *Salmonella* test strains increased with increasing concentrations. However, only decrease in count was noted at 100% concentration (Figure 1).

B. cereus was completely inhibited at concentrations of 25%. Maximum counts for *Staph. aureus* decreased with increasing concentrations. However, no marked difference was noted in initial and final counts at 100% concentration. *L. monocytogenes* growth was affected at 50% and higher concentrations with complete inhibition noted at 100% concentration (Figure 2).

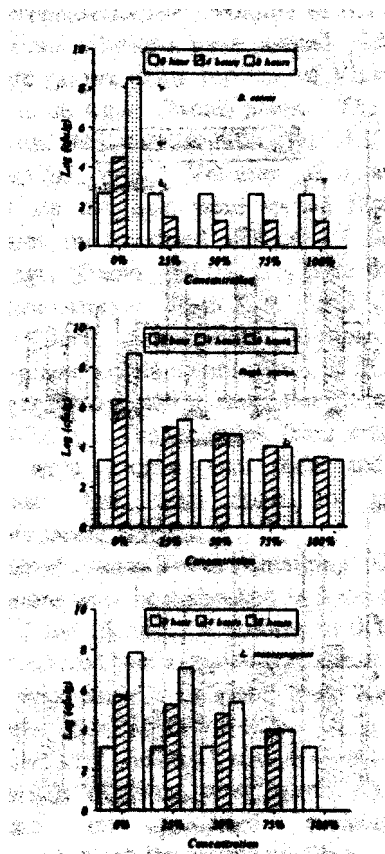


Figure 2: Fate of *B. cereus*, *Staph. aureus*, and *L. monocytogenes* at different concentrations of "chat" juice.

Discussion

The initial microflora of most edible plant leaves comes from air, insects, animals, soil and water. The activity of man, such as cultivation by hand, will introduce and/or distribute microorganisms into ecological niches from which they were previously absent. Finally, the introduction of human and other animal waste material into the water or soil will have an obvious impact on the flora of vegetables (11).

Studies have shown that most of the organisms on fresh vegetables are saprophytes such as coryneforms, lactic acid bacteria, spore formers, coliforms, micrococci and pseudomonads (11). The microflora of "chat" may thus be considered saprophytes that are usually found on many other fresh vegetables.

In comparison to bacterial counts of vegetables upon arrival to a processing plant (12), "chat" leaves have much lower bacterial counts. However, as "chat" leaves are consumed without further cleaning or other treatment, their bacterial load should not be considered acceptable. Considering the fact that "chat" leaves had mean counts of 104 cfu/g for *Staphylococcus*, 47% of which were *Staph. aureus*, the leaves may possibly result in Staphylococcal food poisoning. Enterotoxin production can occur at *Staph. aureus* count of 106 cfu/g (13). The presence of *B. cereus* in "chat" leaves, though at lower levels, may indicate that "chat" can play a role in the transmission of *B. cereus* food poisoning if level of contamination exceeds 106 cfu/g. An outbreak of *B. cereus* food poisoning due to home-nurtured vegetable sprouts has been reported (14).

The "chat" plant is usually cultivated around the homesteads and is highly likely that human and animal wastes are used as fertilizers. Various types of infections such as shigellosis, typhoid fever, cholera, amebiasis (15), outbreaks of salmonellosis (16) and infectious hepatitis (17) are reported

to be caused by raw vegetables where "night soil" was used as fertilizers. Considering the ubiquitous nature of *L. monocytogenes* (18), contamination of "chat" leaves with *L. monocytogenes* is possible. *L. monocytogenes* causes abortion in pregnant women and meningitis and encephalitis particularly in immunocompromised hosts (19).

"Chat" consumption can, thus, be considered hazardous to health and the ingestion of "chat" juice may not have an inhibitory effect on pathogens which may contaminate the leaves.

A decrease in the microbial load of chat leaves may be achieved through a thorough washing of the leaves before chewing.

Acknowledgement

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Brief communication

The molluscicidal effect of the bark of “Bitza” tree (*albizia coriaria*): A local discovery towards the

control of schistosomiasis in the Abay River Valley of Western Ethiopia

Hindjifata Mengesha¹, Hailu Birrie² and Gunnar Gundersen³

Abstract: The extract of the bark of a tree locally known as "Bitza" (*Albizia coriaria*) is traditionally used for fishing by the indigenous population of the western part of the Abay (Blue Nile) River Valley of Ethiopia. The same bark was found to have molluscicidal effect. Laboratory tests showed that 100% of *Biomphalaria* snails die at 50ppm and above when exposed to the water, ethanol or methanol extract of the bark for six hours. Chemical analysis of the extract showed the active ingredients to be tannins which are known to have less active molluscicidal properties than saponins. Although *Albizia coriaria* is not recommended for a largescale use as a molluscicide because of its low potency compared to Endod (*Phytolacca dodecandra*), the discovery demonstrates how local traditional wisdom may form a starting point in the scientific search for other plant molluscicides. [*Ethiop. J. Health Dev.* 1997;11(1):89-92]

Introduction

In developing countries, over 300 million people, mainly children, are infected with schistosomiasis, particularly in rural areas with minimal domestic water supplies, sanitary facilities, or health services (1).

In Ethiopia, too, studies have shown that intestinal schistosomiasis is very widespread and is posing a serious public health problem next to malaria (2). Some of the most affected communities in Ethiopia are those located in the Abay (Blue Nile) River Valley (3).

In realization of the severity of the problem, a community-based control programme was undertaken in Dalati and Agallu Metti areas, both located in the western part of the Ethiopian Abay (Blue Nile) River Valley from 1989 to 1992 (3). The control was based on a primary health care approach using mass chemotherapy and limited focal snail control with niclosamide. Although this programme was successful, sustainability was at stake because of the cost of the drug and niclosamide, both of which were purchased from abroad using hard currency. In view of this, it was sought to resort to Endod (*Phytolacca dodecandra*), whose molluscicidal properties had already been established (4) for use as an alternative molluscicide for a sustained control of the vector snails. As a result, propagation of Endod in the areas was undertaken. The plants, however, failed to survive under the natural agroclimatic conditions of the areas at the time.

The failure of Endod to grow in the areas prompted the first author, who was then a primary health care agent of the project, to look for an alternative plant molluscicide. Utilizing his ethnic

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(Gumuz people who live along the western part of Ethiopia bordering the Sudan) cultural knowledge of locally growing medicinal plants, he tested the molluscicidal properties of the bark of a tree, locally known as "Bitza" and has been used by the communities for fishing for time immemorial. Traditionally, the bark is soaked in shallow water where it is stamped with wooden poles. The extracts result in some kind of intoxication of smaller fishes for some distance downstream. The intoxicated and slowly moving fish are then caught by hand for use as food. Based on this knowledge, the first author performed a crude experiment and observed that snail hosts of *Schistosoma mansoni* died when put in a water containing finely crushed barks of the tree.

Inspired by this observation further laboratory tests were performed using standard procedures. In this short communication, we present the results and discuss how local tradition and ingenuity can form a starting point for scientific investigation of locally available disease control agents.

Methods

The first author collected the bark; ground it; and suspended it in water. To this suspension of unknown concentration, he put *Biomphalaria* snails and observed that all snails died. His preliminary report then led to the collection of the barks of the tree, the branches and leaves, with close-up photographs (Figs. 1 and 2), for identification and further investigations.

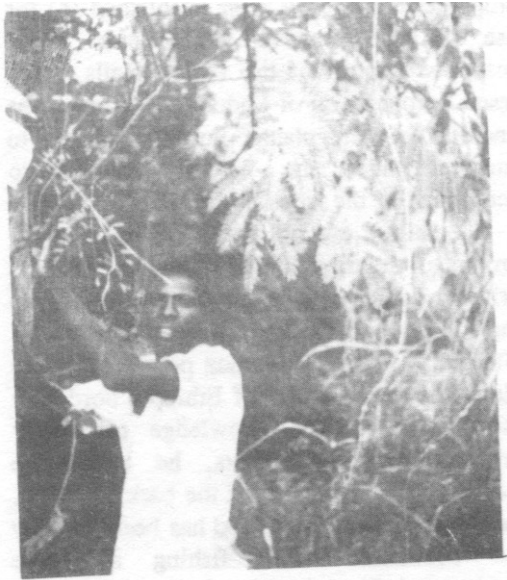


Figure1: The first author with the “bitza” tree (*Albitzia coriaria*).

Testing for molluscicidal property: Part of the bark was sent to the Institute of Pathobiology for further molluscicidal testing. The bark was ground to fine powder and sieved using iron mesh of 250 micron size. The fine powder was weighed and suspended in water to prepare crude concentrations of 50, 100, 200, 400, 800 and 1600 ppm. To each of these concentrations 10 *Biomphalaria* snails were exposed for six hours.



Figure 2: Close-up photograph of the branches of the “bitza” tree

Identification and chemical analysis: Parts of the dry bark, branches and leaves (with close-up photographs, Figs. 1 and 2) were sent to the Institute of Botany at the Agricultural University of Norway, Norway, for botanical identification while part of the bark was sent to the Institute of Organic Chemistry at the Royal School of Pharmacy, Denmark, for chemical analysis. The bark was subjected to ethanol-, methanol- and water extraction according to standard procedures. The resulting extracts were further subjected to chromatographic analyses with the aim of identifying the active ingredients.

Confirmatory testing for molluscicidal property: Parts of the dry bark was sent to the Danish Bilharziasis Laboratory, Denmark, where similar ethanol-, methanol- and water-extracts of the bark were prepared and tested to determine the lowest concentration at which 100% snail mortality (LC100) would be achieved for each extract.

Results

Botanical identification: Based on the morphological characteristics of the bark, branches and leaves, the plant was identified as *Albizia coriaria*.

Chemical analysis: The structures identified by chromatographic examination of the ethanol-, methanol- and water- extracted solutions indicated that the active ingredients were tannins.

Molluscicidal effects: At the Institute of Pathobiology, 100% snail mortality (LC100) of *Biomphalaria pfeifferi* was observed at 100ppm and above when exposed for six or more hours to the crude suspensions of the bark.

In Denmark it was shown that the crude ethanol-, methanol- and water- extracts resulted in 100 % mortality of *Biomphalaria* snails at 50 ppm and above when exposed to the extracts for six hours.

Discussion

Because of the cost of imported synthetic molluscicides, there has for long been a search for locally available plant molluscicides in countries endemic for schistosomiasis. The potential use of plant molluscicides against schistosomiasis was reviewed in the 1983 meeting of the UNDP/World Bank/WHO Special Programme for Research and Training in Tropical Diseases Scientific Working Group on Plant Molluscicides (5). By then, more than 1000 plant species had been

screened for their molluscicidal properties, of which Endod (*P. dodecandra*) was considered as the most promising one (5,6). Community-based trials have demonstrated encouraging results in the control of vector snails of schistosomiasis using Endod (7,8). However, difficulties of growing Endod at varying agroclimatic conditions in Ethiopia (9) necessitate further Endod adaptation studies and/or a search for other plant molluscicides.

The testing of the bark of the "bitza" tree (*A. coriaria*) for molluscicidal properties is part of this continuous search. The most interesting aspect of the present work is that the initiative was taken by a local primary health care agent who is aware of the Endod story. He utilized his native cultural wisdom about traditional medicine in arriving at the present investigation which led to a series of laboratory tests and analyses. Although the bark of the "bitza" tree has a molluscicidal effect at much higher concentration compared to Endod (*P. dodecandra*) which is lethal to 100% of the vector snails at much lower concentrations of 10 ppm and above in the laboratory (1,10) the trial demonstrates how a local ingenuity can form a starting point for scientific investigation of the molluscicidal potential of medicinal plants. In view of the biodiversity of the Ethiopian flora and the diverse and rich cultural wisdom of the Ethiopian people the search need to be encouraged. The low molluscicidal effect of the bark of "bitza" tree (*A. coriria*) is perhaps related to the chemical content of the plant part of which is identified to be tanins instead of saponins. This finding is consistent with a previous report which indicated that the bark of *A. coriaria* contains tannins with molluscicidal activity at concentrations of 100 ppm against *Bulinus truncatus* and *Biomphalaria pfeifferi* (11,12). It has been shown that a bark of the root of another *Albizia* tree, *A. anthelminthica*, contains monodesmodic saponins which have molluscicidal activity against *Biomphalaria glabrata* (13,14). However, no saponin-like chemical structure was found in the bark of the "bitza" tree in the present study.

Even if potent, in principle, the use of a bark of a tree as a molluscicide may not be sound both environmentally and economically. However, the fact that the "bitza" bark is still used by the local population in the western part of the Abay (Blue Nile) River Valley of Ethiopia for fishing purposes makes it an important plant to note. An indirect effect of such practices may contribute to a focal snail control in the parts of the rivers where it is applied. It must be noted that dry leaves of *Ambrosia maritima* tree have been used for the control of schistosome intermediate hosts at much higher concentrations of 150-300 ppm in the Senegal River Basin, Senegal (15).

Until other local plant molluscicides are available the recently selected potent variety of Endod (Type-44) needs to be adopted at varying agro-climatic zones of Ethiopia for use in schistosomiasis control by the endemic communities on a self-help basis. Furthermore, the need for identifying other varieties of Endod and/or improving the existing ones, through genetic manipulation, for adaptation and propagation under varying agroclimatic conditions of Ethiopia cannot be overemphasized.

Acknowledgement

We want to thank P. Furu, Danish Bilharziasis Laboratory, Copenhagen, for screening the molluscicidal properties and S. Thilborg, Royal Danish School of Pharmacy, Copenhagen, for screening of chemical contents of the "bitza" bark. Dr. J. Kielland-Lund, Institute of Botany, Agricultural University of Norway, helped the identification of the leaves and bark of the "bitza" tree.

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Letter to the Editor

Findings on the Eye disease connected with Herpes Zoster at the Balcha Hospital

Doctors at the Balcha Hospital have found severe cases of the eye disease connected with Herpes Zoster, that can result in complete blindness or reduced sight if the disease is not treated. Herpes Zoster is very often connected with HIV virus. Disease of the eye with Herpes Zoster can be classified as

1. Herpes Zoster of the eyelids
2. Conjunctivitis
3. Keratitis
4. Uveitis
5. Keratouveitis
6. Combination of two or more of the above

Forty six patients with Herpes Zoster have been examined between 1993-96 and only three of these are found to be free from Herpes Zoster of the eyelids. However, the three patients had Keratouveitis connected with Herpes Zoster of skin of other parts of the body. Intensive course of local and general treatment has been performed on these patients and the recovery has been found to have enhanced three to four times. No case of blindness has been observed to have recurred after the treatment. Nevertheless three patients out of eight who have showed up at our hospital have been found to be completely blind as a result of having no treatment at all. No complication has been observed on the patients treated for the disease so far.

Korneev Yuri M.
Ophtalmologist
11th December 1996

Notice Board

Announcement

The 8th Annual conference of the Ethiopian Public Health Association will be held 6-8 November 1997. The venue will be Medical Faculty Auditorium, Tikur Anbessa Hospital Addis Ababa.

The deadline for submission of Abstracts to be considered for presentation at the conference is June 30, 1997.

Training Opportunities

1. Effective leadership the latest skills and concepts, 5-30 May, 1997, Boston, Massachusetts, USA
2. Executive Course in Health Financing and Sustainability , 9-27 June 1997, Boston, Massachusetts, USA
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