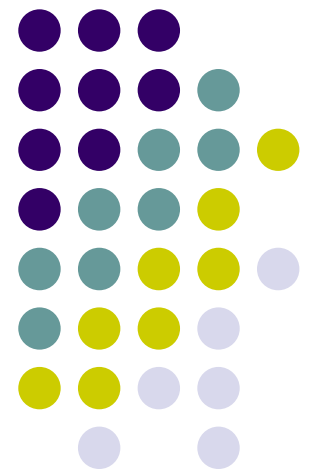


37th Union World Conference on Lung Health
Strengthening human resources for better lung health

The challenge of TB laboratory diagnosis in the HIV-infected

Does bleach concentration of sputum improve the sensitivity of smear-microscopy in HIV-positive patients?

Sara Eyangoh
Centre Pasteur du Cameroun

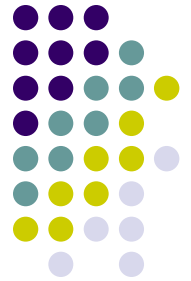


TB diagnosis using Z-N smear-microscopy



- Low and variable sensitivity of Z-N methods well known (20-60%)
- ZN an old technique since 1883
- Remains the main tool for TB diagnosis /identification of AFB
 - Simplicity
 - Rapidity
 - Low cost
 - Feasibility on the field
 - High specificity for *M. tuberculosis* in TB endemic countries

Factors of low sensitivity of ZN



- Sample collection
 - Is the quantity enough?
 - Is the quality suitable?
- Technical aspects
 - are the standard procedures for smear preparation and staining respected?
 - is the microscope of good quality?
- Person carrying out the test
 - Motivation and training
 - Time of smear reading



Problem of TB diagnosis in the HIV-infected



- TB is the commonest HIV related disease
- Lower bacillary load in HIV+ than in HIV -
- This make TB harder to diagnose in HIV +



Diagnostic value eroded due to increasing number of HIV-related smear negative TB



Necessity to improve the sensitivity

Methods use for improvement



- Smear microscopy improve by liquefaction with chemicals reagent and concentration by centrifugation or sedimentation
 - detergents (C18-Carboxypropylbetaine/ Tween 80)
 - Chitin
 - NaOH
 - Acetyl-Lcysteine + NaOH
 - NaOCl known as bleach household
- Relevant systematic review:
 - Steingart & coll. 2006. Lancet.infect.Dis
 - Ängeby & coll. 2004. Int.J.Tuberc.Lung.Dis.

The bleach concentration methods



- Many advantages for household bleach
 - Cheap, available everywhere
 - Reduce biohazards associated with smear preparation
 - turning smear classified « scanty » into +
- The sensitivity gained varied between (0%- >100%)
- Few studies considered the sensitivity in the HIV infected



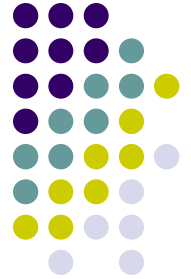
Does bleach concentration methods improve the sensitivity of smear microscopy in the HIV infected?

Study design in Cameroon



- Settings
 - Diagnostic centre at Jamot hospital, Yaoundé
 - Mycobacteria Laboratory, Pasteur Institute, Yaoundé
- Subjects
 - All consecutive suspects sent at Jamot diagnostic center without age and sex consideration
- Sample
 - 1 specimen of morning sputum/suspect; any volume
 - 1000 sputum from as many suspect sent at Mycobacteria Laboratory with ZN result of Jamot diagnostic center
- Funding: USAID

objectives



Evaluation of sputum concentration for the diagnosis of TB

Comparison between both the direct and Concentrated smear by Z-N and fluorescence

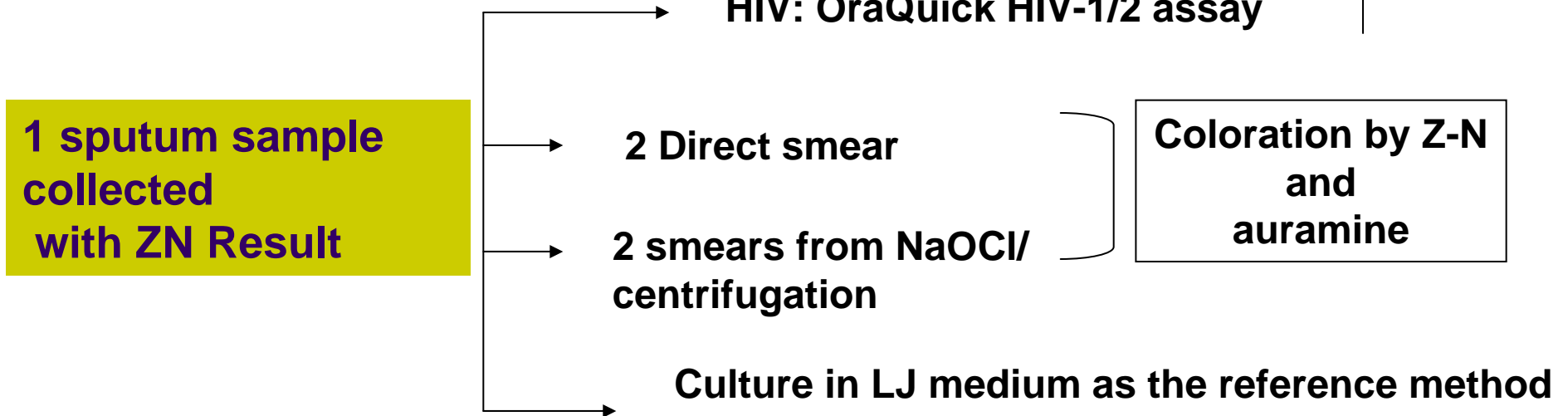
comparison of methods Sensitivity between non infected and infected HIV patients

Comparison of results obtained in Diagnostic centre and the reference laboratory

Methods

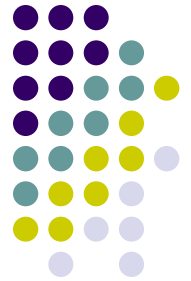


Specimen examination



- **Blind reading: 2 technicians; HIV-status hidden**
- **Direct smears and processed smears read separately**
- **Smears results interpreted independently of culture**
- **Study was approved by Cameroon ethical committee**

Patient characteristics (n=940)



- Masculine sex : n= 506 (54%)
- Feminine sex : n= 434 (46%)
- Predominant active-life period: 37,5 ± 15 years
- HIV (+): n= 422 (45 %)
- HIV(-): n= 513 (54,5%)
- HIV Undetermined: n=5 (0.5%)
- Positive culture: 419 (44 %)
- TB +VIH: 184 (43%)

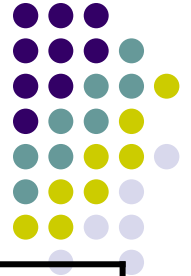
Sensitivity and specificity of tests



test		Culture + (419)	Culture – (521)
ZN	positive	n=278 Se=66.3%	n=3
	negative	n= 141	n=518 Sp=99.4%
ZN + NaOCl	positive	n= 308 Se= 73.5%	n=2
	negative	n= 111	n= 519 Sp=99.6%

Sputum treated with bleach increased ZN sensitivity significantly

Sensibility tests considering HIV status



		HIV+ (422)		HIV- (513)	
		Culture+ (184)	Culture - (241)	Culture + (235)	Culture - (280)
ZN	positive	84 Se= 46.2%	0	193 Se= 82%	3
	negative	99	N=241 Sp=99.9%	N=40	N= 277
ZN+ NaOCl	positive	101 Se= 54.9	1	207 Se = 88%	1
	negative	83	240	26	279

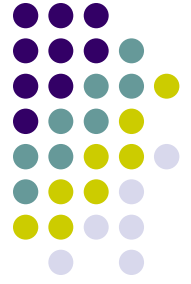
Significant Increased proportion of positive tests with bleach in HIV+ as well as HIV-negative compared to direct Z-N

Results



- Sensitivity of smear microscopy is low in HIV+ (46% vs 82%)
- In HIV +: sensibility gained is 8.2%
- In HIV- :sensibility gained is 6%
- Scanty proportion by HIV status
 - Z-N (49): VIH- (26) vs VIH+ (23)
 - ZN+NaOCl (31) : VIH- (13) vs VIH+ (18)
- Same results with fluorescence compare to ZN

Conclusion



- Bleach processing increases the sensitivity in HIV+ compared with direct microscopy
- But the increase is modest in our study
- Workload is high, but reading more ease
- We haven't considered the volume and the quality of sputum



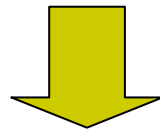
Recommendations



- Other studies are needed for the determination of appropriate use of sputum processing methods:
 - Effect of different processing method under controlled conditions
 - blinded multicenter studies in setting of high and low HIV prevalence



**strengthening the quality management
of laboratory to improve the reliability of diagnosis**



Confirm performance of processed method in settings
and determine case-finding,
Cost, workload and programmatic impact