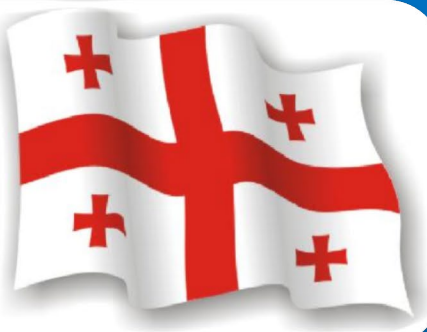
NATIONAL CENTER FOR TUBERCULOSIS AND LUNG DISEASES



# Detection of MTB in suspected TB patients BAL Fluid by using Genotype®MTBDRplus assay.

D. Tchkonia, K. Vacharadze,
N. Bablishvili, N. Shubladze, N. Tadumadze
Tbilisi, Georgia.

# JOINT PRESS RELEASE

## New ECDC-WHO report: tuberculosis on the retreat in Europe. Concerns about drug-resistant TB and treatment failure.

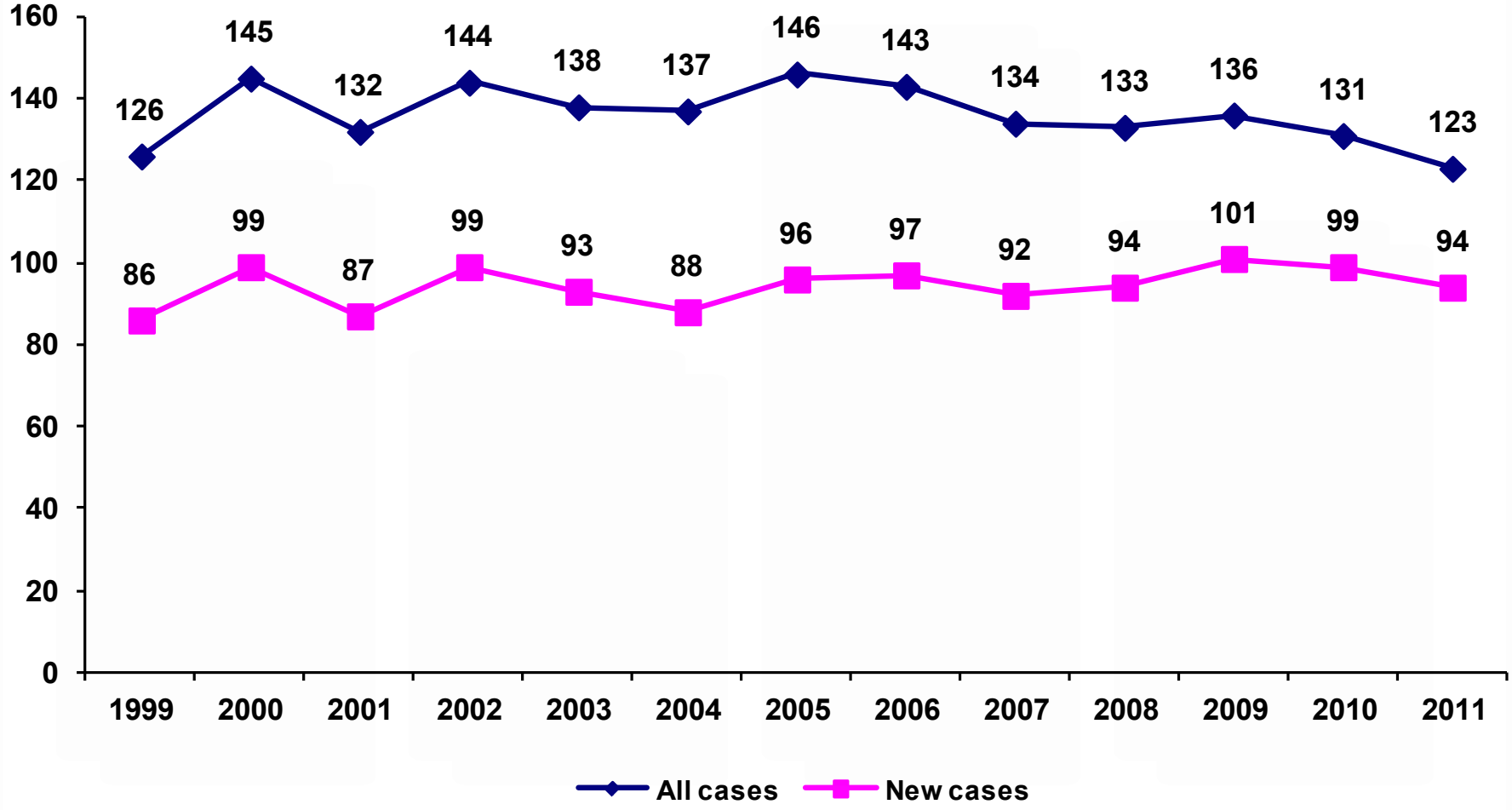
Stockholm/Copenhagen, 19 March 2012

"Our Region has the lowest TB treatment success rate and the highest M/XDR TB rate in the world," says **Zsuzsanna Jakab, WHO Regional Director for Europe**. "TB has not spared any country in our Region, but has hit hardest in the 18 high priority TB countries mainly in the eastern part of the Region[1]. This is something we must address as a matter of urgency. Special attention should be paid to timely diagnosis and adequate treatment of all types of the disease with particular attention to vulnerable populations."

[1] Armenia, Azerbaijan, Belarus, Bulgaria, Estonia, Georgia, Kazakhstan, Kyrgyzstan, Latvia, Lithuania, Moldova, Romania, Russian Federation, Tajikistan, Turkey, Turkmenistan, Ukraine, Uzbekistan

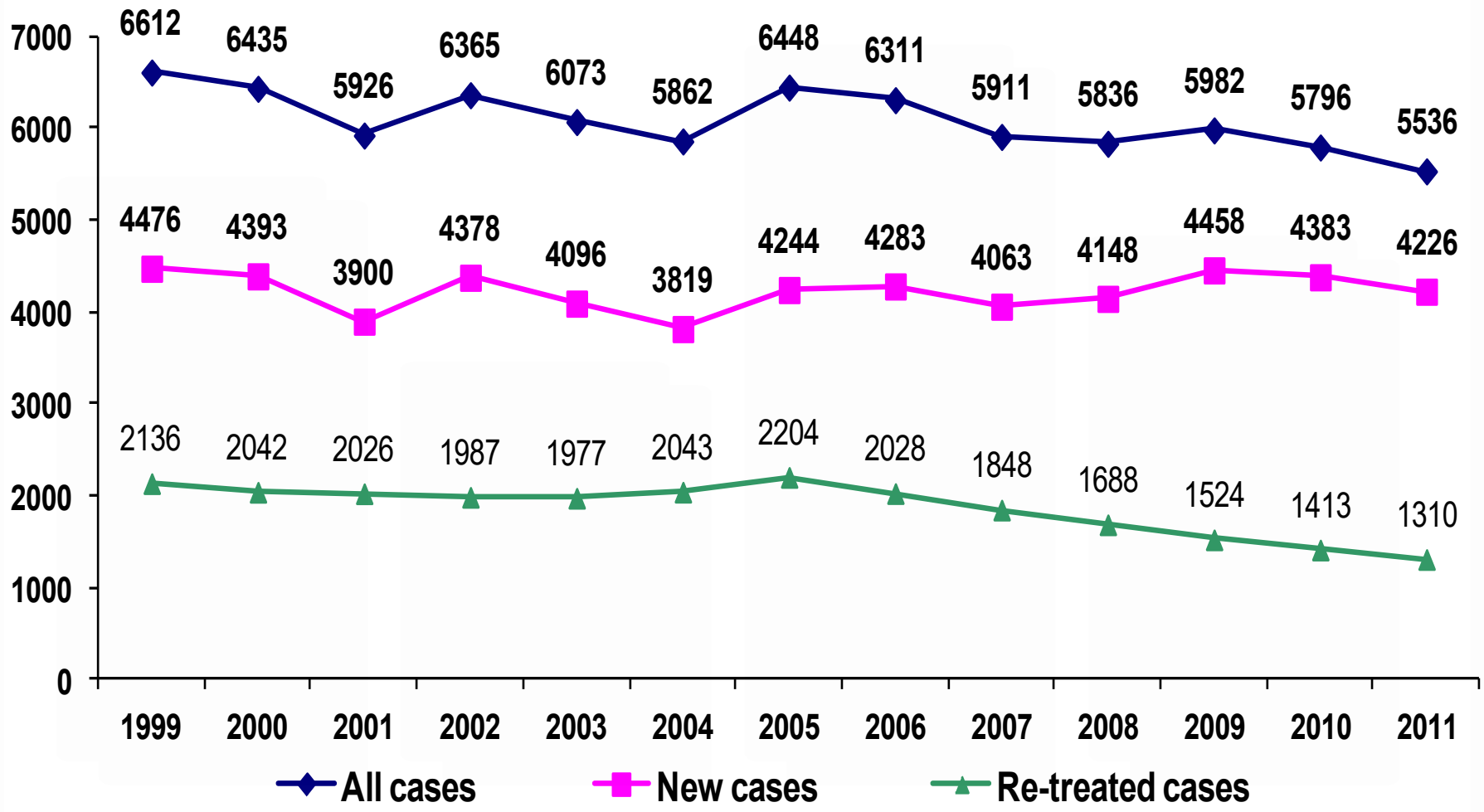
# Incidence of tuberculosis in Georgia

## (per 100,000 people)

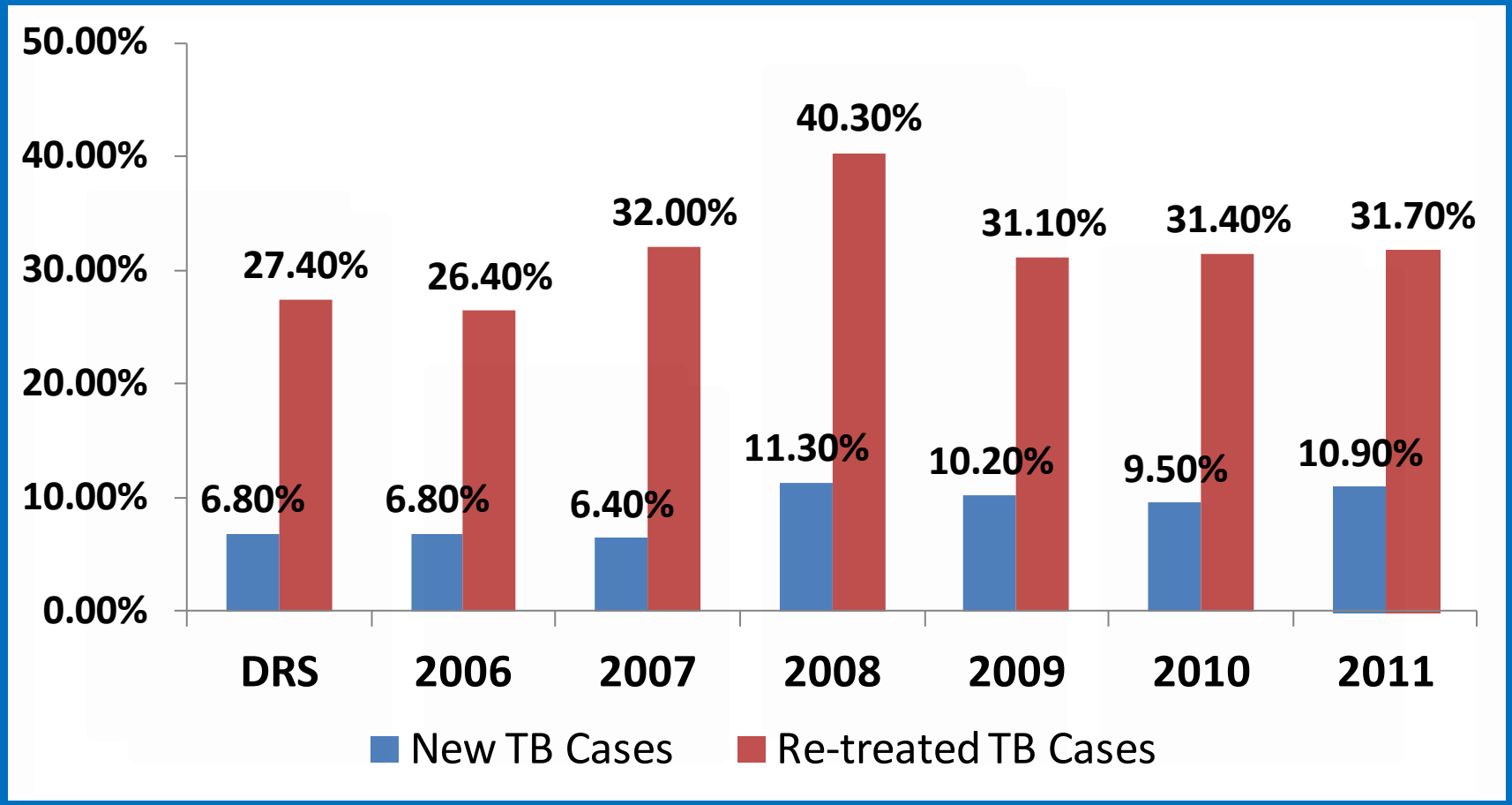


# Incidence of tuberculosis in Georgia

## (In absolute numbers)



# Surveillance of drug resistance in tuberculosis MDR-TB increased: 2009: 369; 2010: 359; 2011: 475



# Drug Susceptibility Testing

**Solid Media**
Löwenstein-Jensen
(Middlebrook)

→ 3 - 4 weeks

**Liquid Media**
BACTEC 460 TB
MGIT

→ 7 - 10 days

**Molecular based Methods**
InnoLipa
GenoTypeMTBDR
Xpert MTB

→ Hours – 1day

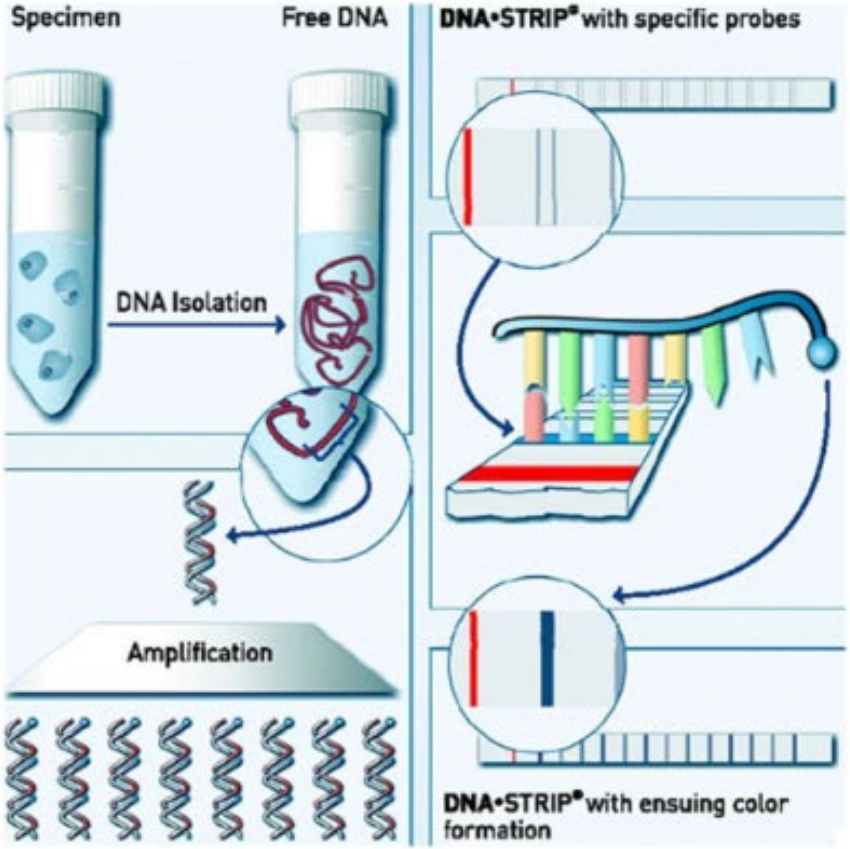
# PCR-based Genotype®MTBDRplus assay

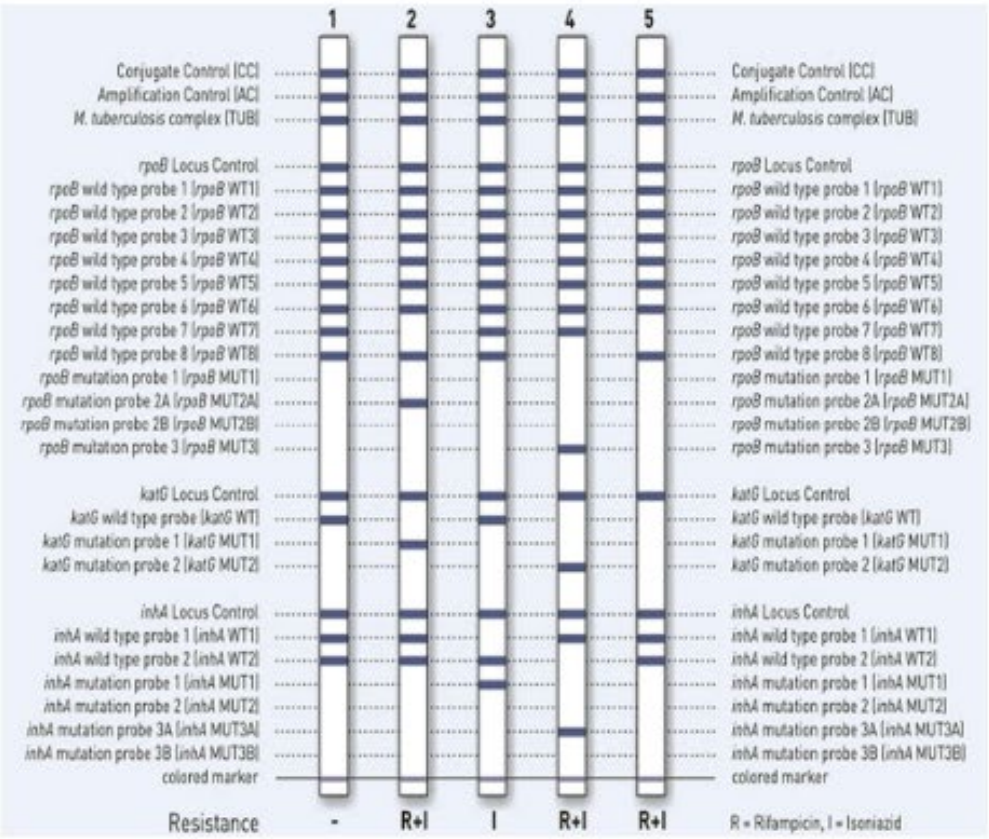
- Drug susceptibility testing (DST) by conventional culture for Mycobacteria is slow and takes several weeks (on average 4 weeks and maximum incubation of 9 weeks).
- Overall performance of MTBDR*plus* assay is superior to conventional culture/DST: speed, interpretable results
- Results in 5 hrs
- Easy to perform, suitable for routine lab

**WHO recommends molecular MTBDR*plus* for rapid diagnosis of MDR-TB in high burden, low-income setting**

# GenoType® MTBDR*plus* Procedure

Specimen → DNA extraction → PCR → Hybridization → Color reaction



A total of 53 BALF samples were examined; 23 out-patients; 30 in-patients; age from 18 to 76.

The Genotype®MTBDRplus system was applied to screen the strains for the rapid MTBC detection and presence of rpoB (S531L, H526D, H526Y and D516V), katG (S315T) and inhA promoter region (C15T and A16G) mutations. The target loci were amplified by PCR and then hybridized with the respective site-specific and wild type (control) probes. Out of 53 patients 18 (34%) didn't produce sputum and 35 (66%) were sputum-smear negatives.

Interestingly, that 5 patients without sputum production began to produce after BALF procedure.

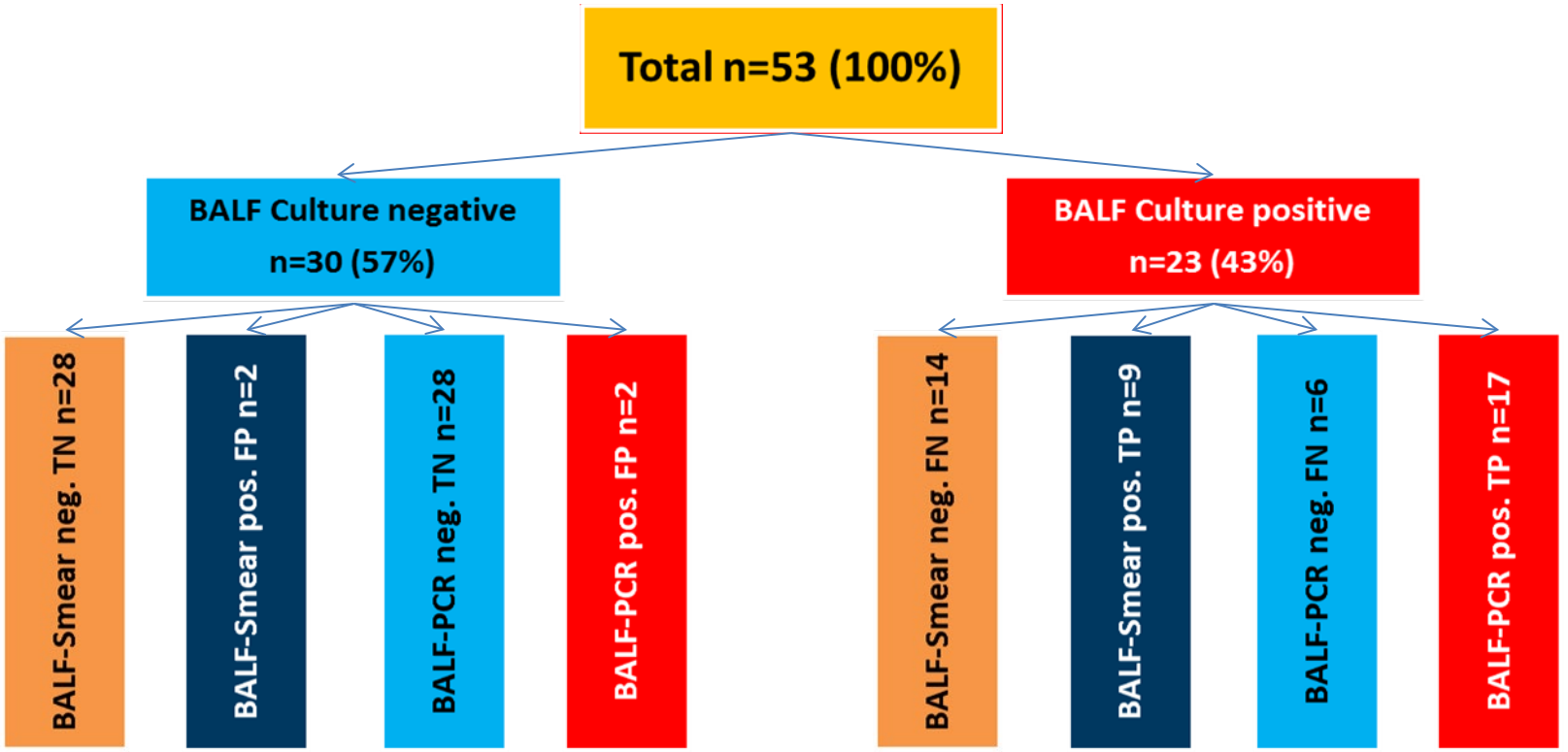
| N | Gender | Age | Dep. | BALF-PCR | BALF-Smear | BALF-Culture |
|---|---|---|---|---|---|---|
| 1 | female | 42 | out-p. | TB-complex -/R | Neg | Neg |
| 2 | female | 62 | out-p. | TB-complex R/R | - | Pos |
| 3 | male | 32 | in-p. | Neg | Neg | Neg |
| 4 | male | 61 | out-p. | Neg | Neg | Neg |
| 5 | male | 64 | out-p. | Neg | Neg | Neg |
| 6 | female | 23 | out-p. | TB-complex S/S | Neg | Pos |
| 7 | male | 53 | out-p. | Neg | Neg | Neg |
| 8 | male | 74 | in-p. | Neg | Neg | Neg |
| 9 | male | 52 | in-p. | TB-complex S/S | Neg | Pos |
| 10 | male | 47 | in-p. | TB-complex R-R | Pos | Pos |
| 11 | female | 18 | in-p. | Neg | Neg | Pos |
| 12 | male | 37 | in-p. | Neg | Neg | Neg |
| 13 | female | 28 | in-p. | Neg | Neg | Neg |
| 14 | male | 36 | in-p. | Neg | Neg | Pos |
| 15 | female | 37 | out-p. | TB-complex S/S | Neg | Pos |
| 16 | male | 72 | out-p. | Neg | Neg | Neg |
| 17 | female | 76 | in-p. | TB-complex S/S | Neg | Pos |
| 18 | female | 68 | in-p. | Neg | Neg | Neg |
| 19 | male | 31 | out-p. | Neg | Neg | Neg |
| 20 | male | 38 | in-p. | Neg | Neg | Neg |
| 21 | male | 68 | in-p. | Neg | Neg | Neg |
| 22 | female | 29 | in-p. | Neg | Neg | Neg |
| 23 | female | 19 | in-p. | Neg | Neg | Neg |
| 24 | male | 68 | out-p. | TB-complex S/S | Neg | Pos |
| 25 | female | 60 | in-p. | Neg | Neg | Neg |
| 26 | female | 28 | in-p. | TB-complex S/S | Pos | Pos |
| 27 | male | 58 | in-p. | Neg | Neg | Pos |
| 28 | female | 50 | out-p. | Neg | Neg | Neg |
| 29 | female | 61 | in-p. | Neg | Neg | Neg |
| 30 | male | 19 | in-p. | Neg | Neg | Neg |
| 31 | female | 31 | in-p. | Neg | Neg | Neg |
| 32 | female | 40 | out-p. | TB-complex S/S | Pos | Neg |
| 33 | female | 47 | out-p. | Neg | Neg | Neg |
| 34 | male | 56 | in-p. | Neg | Neg | Neg |
| 35 | male | 25 | out-p. | Neg | Neg | Neg |
| 36 | male | 18 | out-p. | TB-complex S/S | Pos | Pos |
| 37 | female | 54 | out-p. | Neg | Neg | Neg |
| 38 | male | 61 | in-p. | TB-complex S/ | Neg | Pos |
| 39 | male | 68 | out-p. | Neg | Neg | Neg |
| 40 | male | 49 | in-p. | Neg | Neg | Neg |
| 41 | female | 23 | in-p. | TB-complex S/S | Pos | Pos |
| 42 | male | 75 | in-p. | Neg | Neg | Pos |
| 43 | male | 23 | in-p. | TB-complex S/S | Pos | Pos |
| 44 | male | 58 | out-p. | TB-complex S/S | Pos | Pos |
| 45 | male | 42 | out-p. | Neg | Neg | Neg |
| 46 | female | 41 | in-p. | Neg | Neg | Neg |
| 47 | male | 45 | in-p. | Neg | Pos | Neg |
| 48 | male | 26 | out-p. | TB-complex S/S | Pos | Pos |
| 49 | male | 24 | in-p. | Neg | Neg | Pos |
| 50 | female | 24 | out-p. | TB-complex S/S | Pos | Pos |
| 51 | female | 25 | out-p. | TB-complex S/S | Neg | Pos |
| 52 | female | 50 | out-p. | TB-complex S/S | Pos | Pos |
| 53 | male | 27 | in-p. | Neg | Neg | Pos |

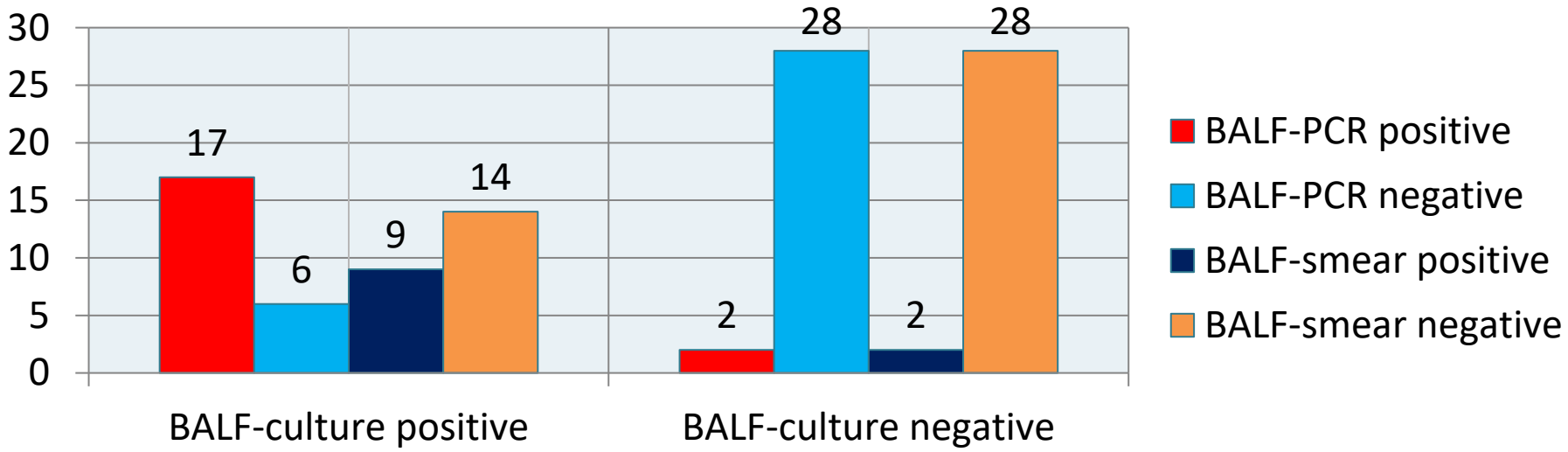
# Culture positive cases

23 (43%) were BALF culture-positive; 9 (36%) had a positive BALF-smear, and 17 (74%) were BALF-PCR positive by Genotype®MTBDR plus assay.

Patients, diagnosed by TB were admitted to us with the diffferent preliminary diagnosis.

| N | Gender | Age | Dep. | BALF-PCR | BALF-Smear | BALF-Culture | Preliminary Diagnosis |
|---|---|---|---|---|---|---|---|
| 1 | female | 62 | out-p. | TB-complex R/R | Neg | Pos | Asthma |
| 2 | female | 23 | out-p. | TB-complex S/S | Neg | Pos | Examinee |
| 3 | male | 52 | in-p. | TB-complex S/S | Neg | Pos | Pleuritis |
| 4 | male | 47 | in-p. | TB-complex R/R | Pos | Pos | TB in 2003 |
| 5 | female | 18 | in-p. | Neg | Neg | Pos | Pneumonia |
| 6 | male | 36 | in-p. | Neg | Neg | Pos | Examinee |
| 7 | female | 37 | out-p. | TB-complex S/S | Neg | Pos | Examinee |
| 8 | female | 76 | in-p. | TB-complex S/S | Neg | Pos | TB in 1956 |
| 9 | male | 68 | out-p. | TB-complex S/S | Neg | Pos | Abscess |
| 10 | female | 28 | in-p. | TB-complex S/S | Pos | Pos | TB contact |
| 11 | male | 58 | in-p. | Neg | Neg | Pos | TB in 1980 (prison) |
| 12 | male | 18 | out-p. | TB-complex S/S | Pos | Pos | Lymphoma |
| 13 | male | 61 | in-p. | TB-complex S/ | Neg | Pos | Cr? TB contact |
| 14 | female | 23 | in-p. | TB-complex S/S | Pos | Pos | Examinee |
| 15 | male | 75 | in-p. | Neg | Neg | Pos | COPD. Cr? |
| 16 | male | 23 | in-p. | TB-complex S/S | Pos | Pos | Hemoptysis, Pulmonary Infarction? |
| 17 | male | 58 | out-p. | TB-complex S/S | Pos | Pos | Examinee |
| 18 | male | 26 | out-p. | TB-complex S/S | Pos | Pos | Examinee |
| 19 | male | 24 | in-p. | Neg | Neg | Pos | TB contact (prison), Hemoptysis |
| 20 | female | 24 | out-p. | TB-complex S/S | Pos | Pos | Examinee |
| 21 | female | 25 | out-p. | TB-complex S/S | Neg | Pos | Examinee |
| 22 | female | 50 | out-p. | TB-complex S/S | Pos | Pos | Examinee |
| 23 | male | 27 | in-p. | Neg | Neg | Pos | Pleuritis |

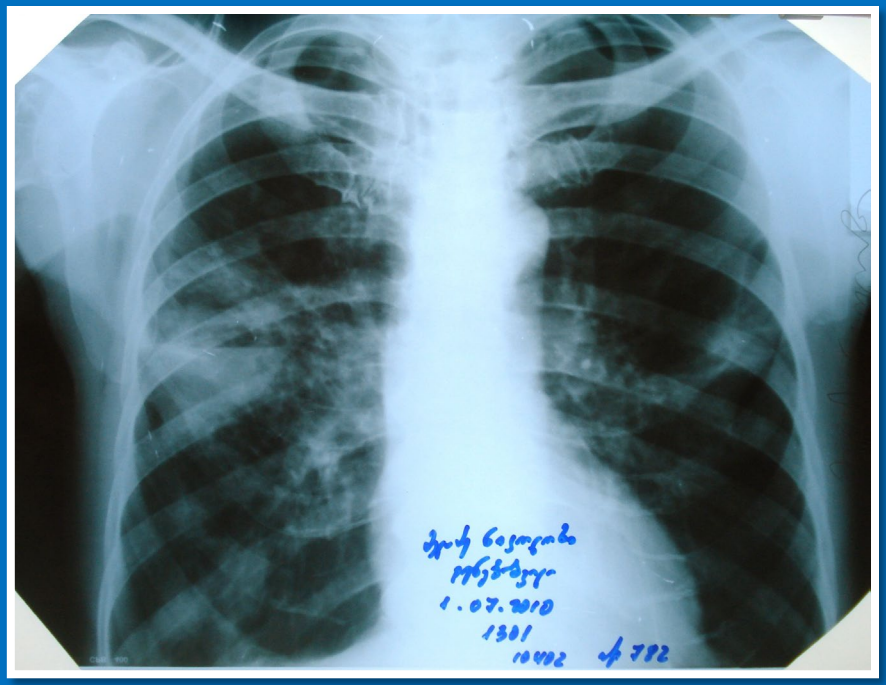

Total n=53 (100%)
BALF Culture negative
n=30 (57%)
BALF Culture positive
n=23 (43%)
BALF-Smear neg. TN n=28
BALF-Smear pos. FP n=2
BALF-PCR neg. TN n=28
BALF-PCR pos. FP n=2
BALF-Smear neg. FN n=14
BALF-Smear pos. TP n=9
BALF-PCR neg. FN n=6
BALF-PCR pos. TP n=17

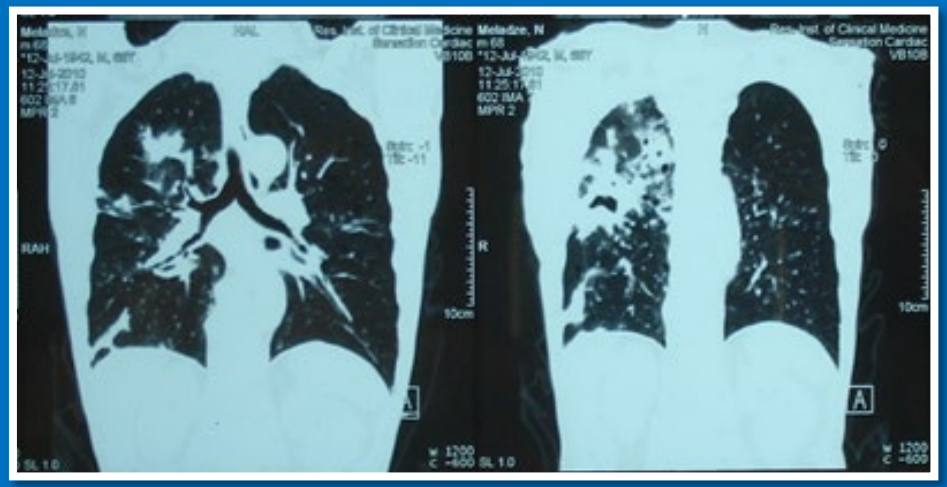

30
25
20
15
10
5
0
17
6
9
14
2
28
2
28
BALF-culture positive
BALF-culture negative
BALF-PCR positive
BALF-PCR negative
BALF-smear positive
BALF-smear negative

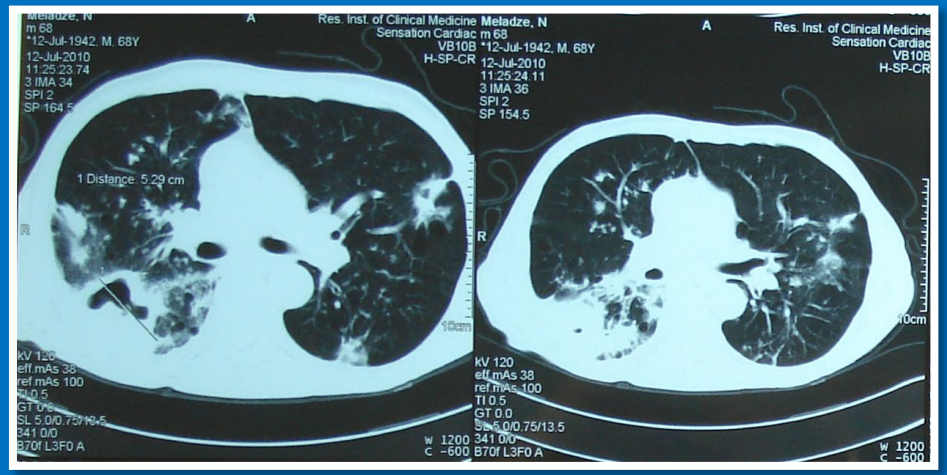
# Comparison between BALF-Smear and BALF-PCR

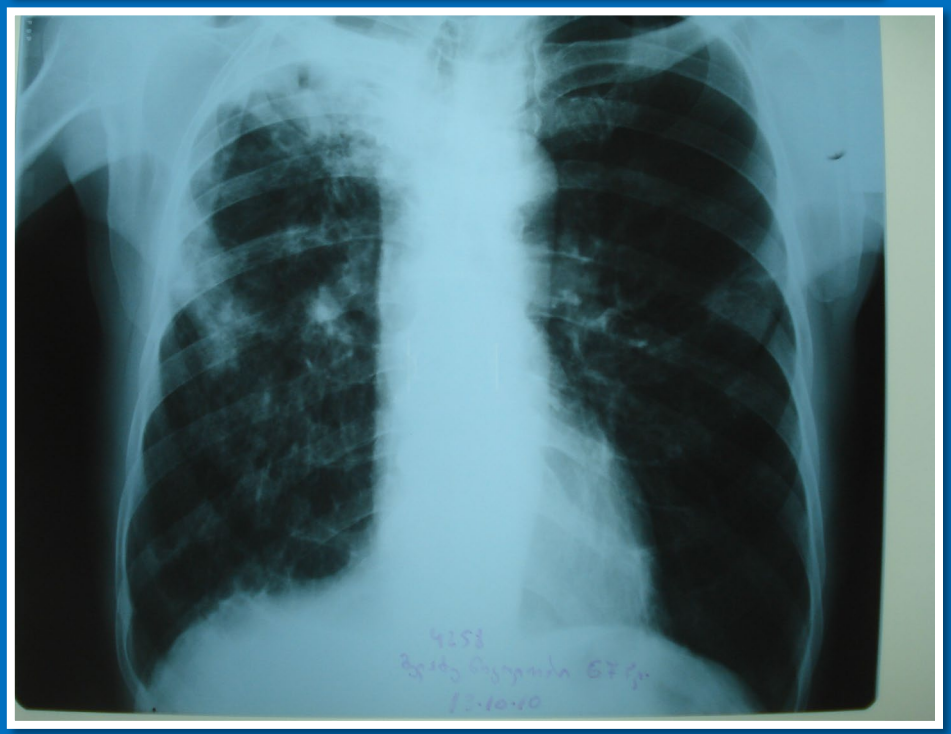
| | Sensitivity | Specificity | PPV | NPV |
|---|---|---|---|---|
| BALF-PCR | 74% | 93% | 89% | 82% |
| BALF-smear | 39% | 93% | 81% | 67% |

Out-patient; 68 years old male; 20 day medical history; fever 38-39 C; WBC 11,0X10$^3$ K/uL; ESR 62 mm/hr; weakness; weight loss; never suffered from tuberculosis; had no TB contact; was treated with antibiotics.

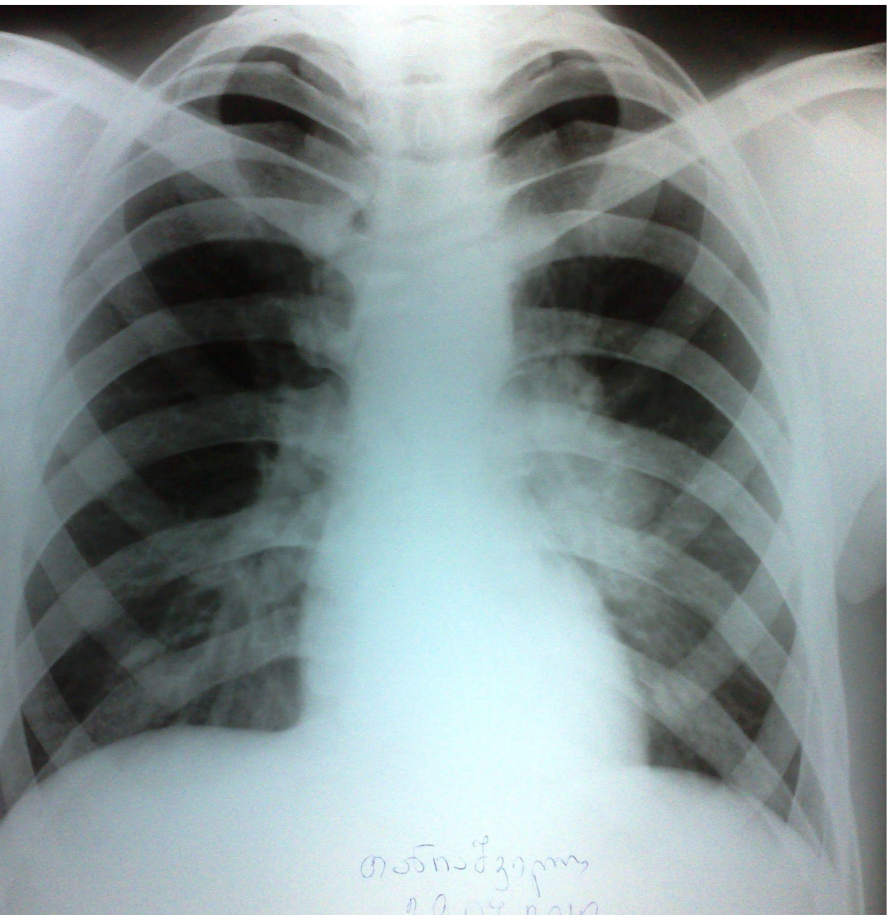


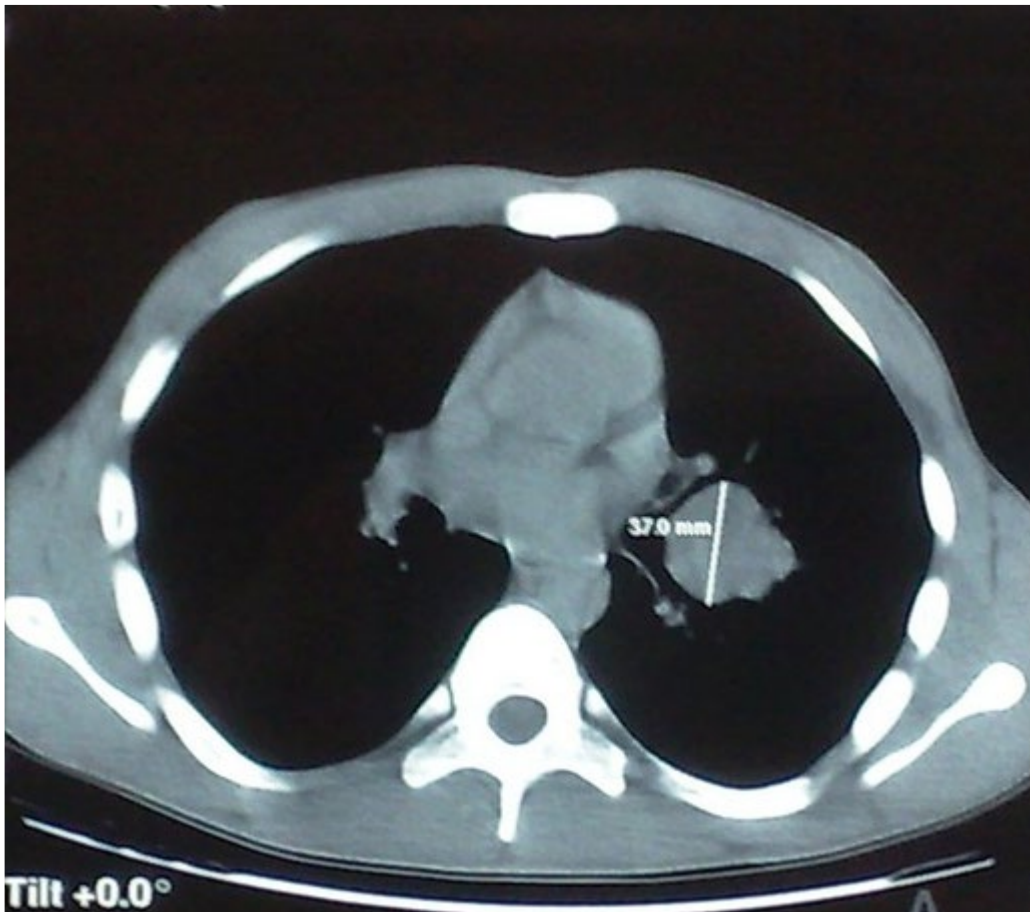


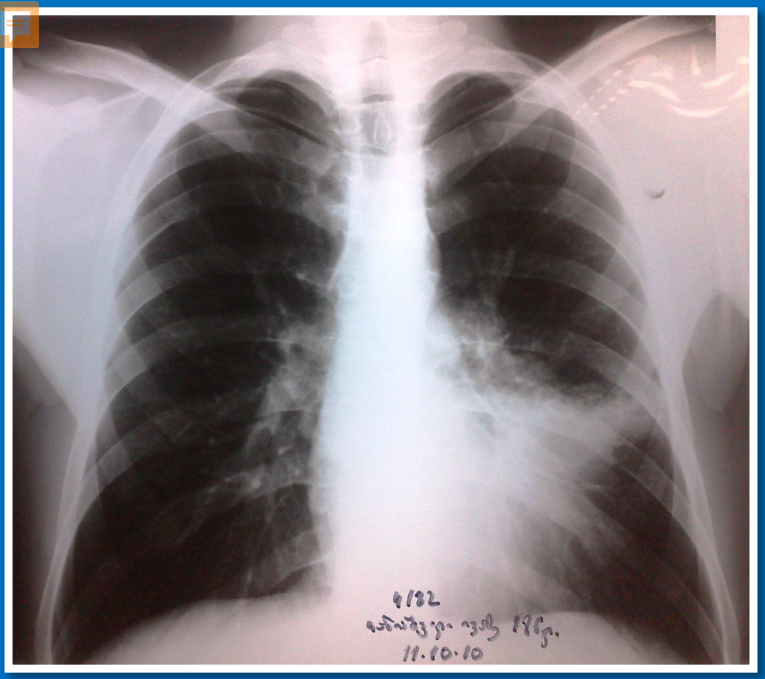


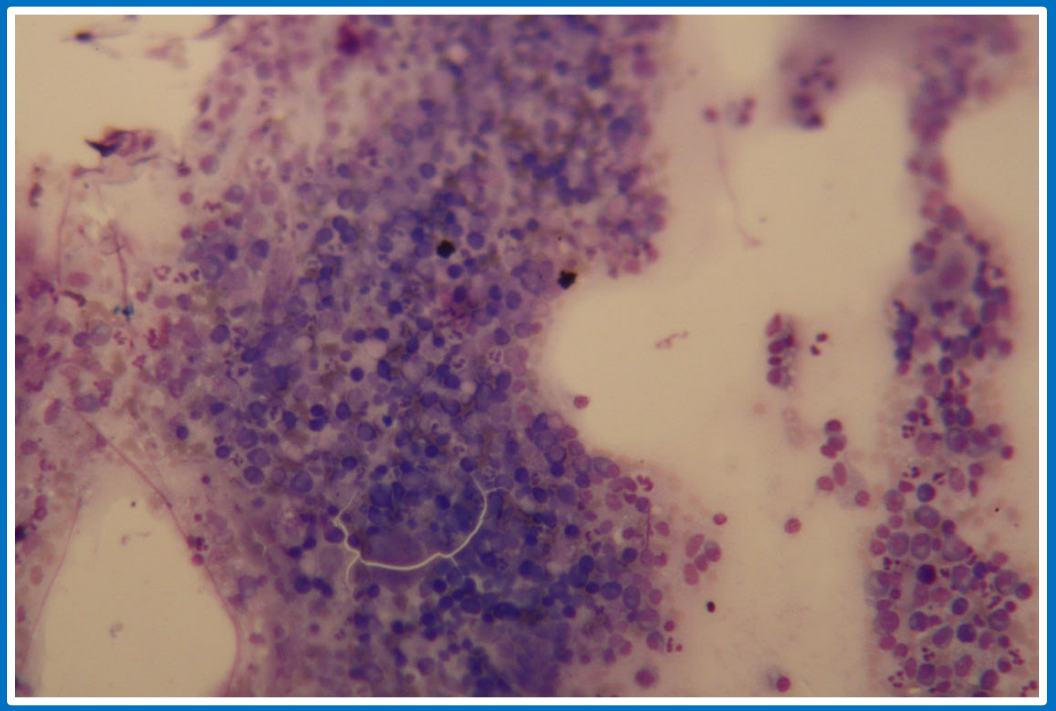



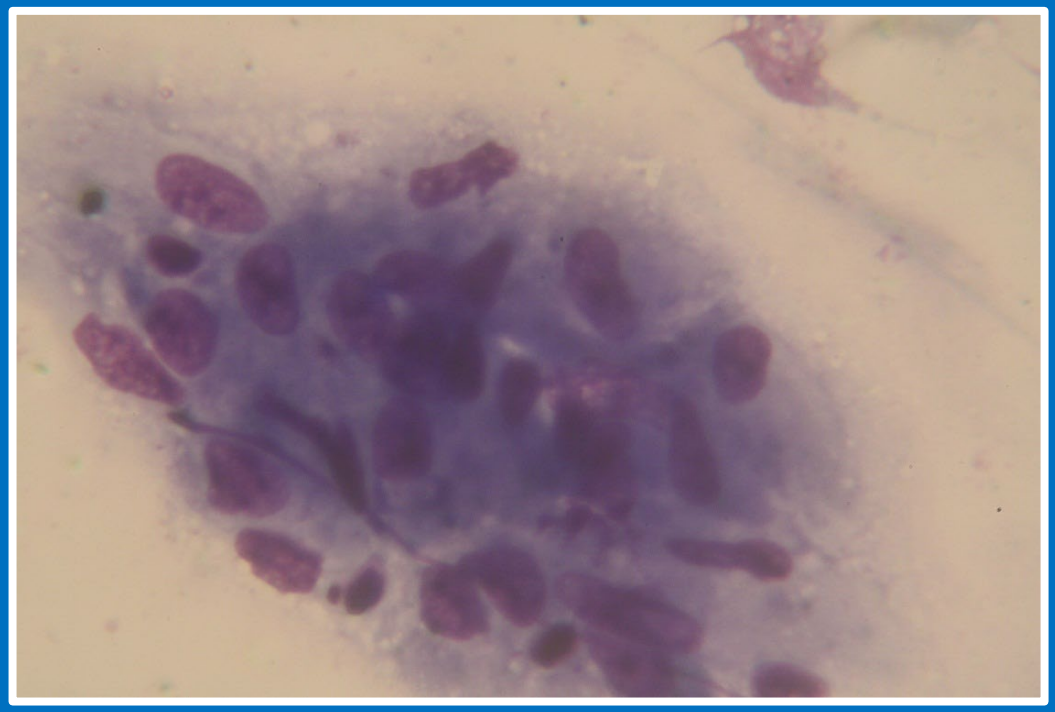
Out-patient; 18 years old male ; 1 month of medical history;  without fever; weakness; pain in the left chest during a deep breath; never suffered from tuberculosis; had no TB contact; was treated by antibiotics.

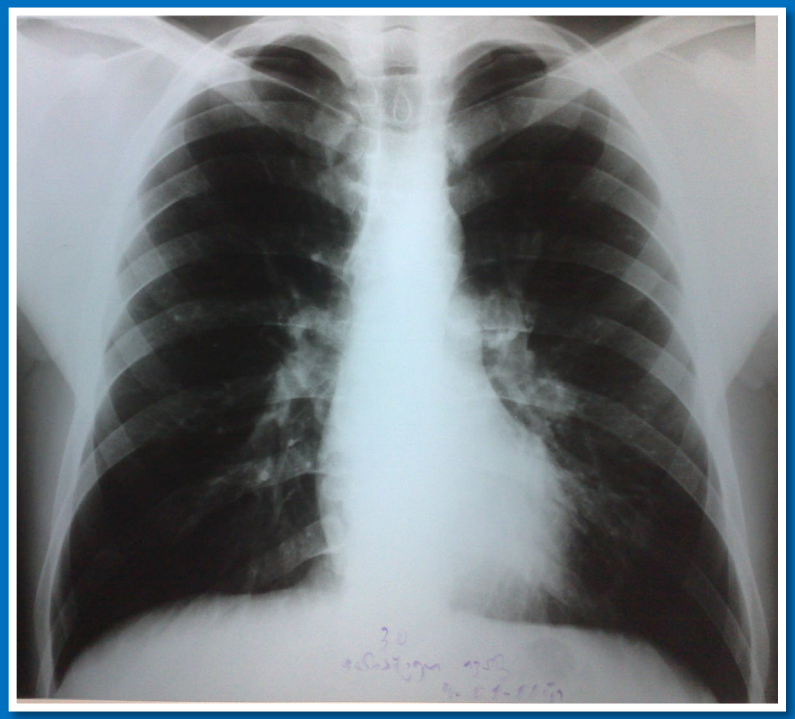












# CONCLUSION

- BAL procedure contributed the appearance of sputum.
- Combined use of BALF-smear and MTB complex-MTBDRplus assay has a good diagnostic yield in complex cases with suspected TB.
- PCR-based Genotype®MTBDRplus assay may be suggested for wide use.
- The future of TB diagnosis remains in the application of new molecular techniques but The gold-standard is the culture, and the other methods have to be considered and interpreted as complementary diagnostic methods.

# Bronchoscopic techniques performed for the first time in Georgia.

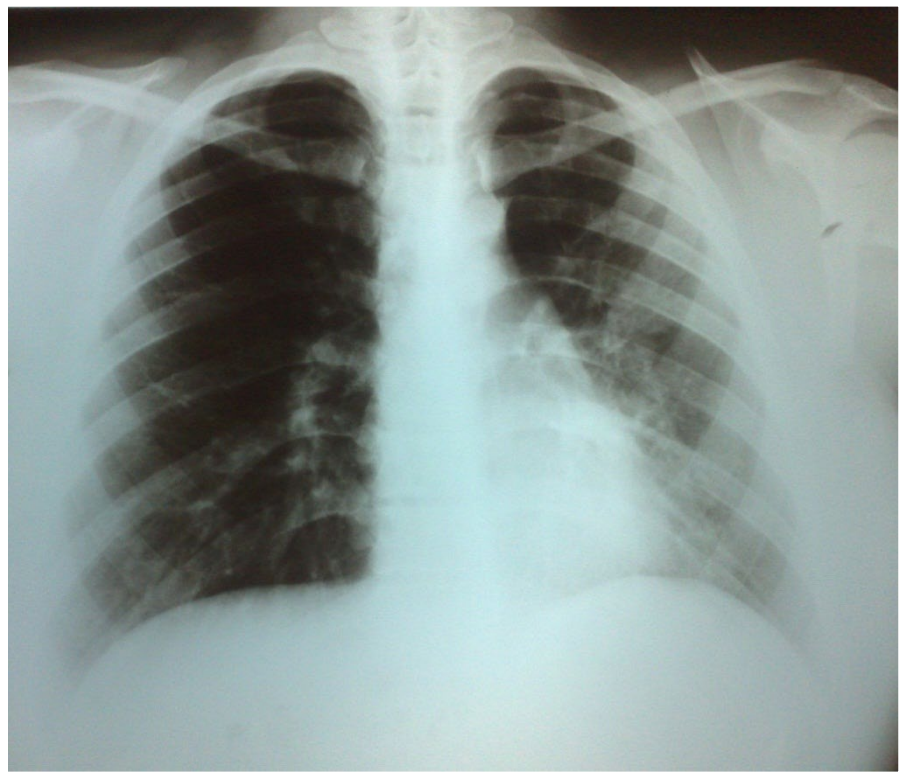
## Airway Stenting

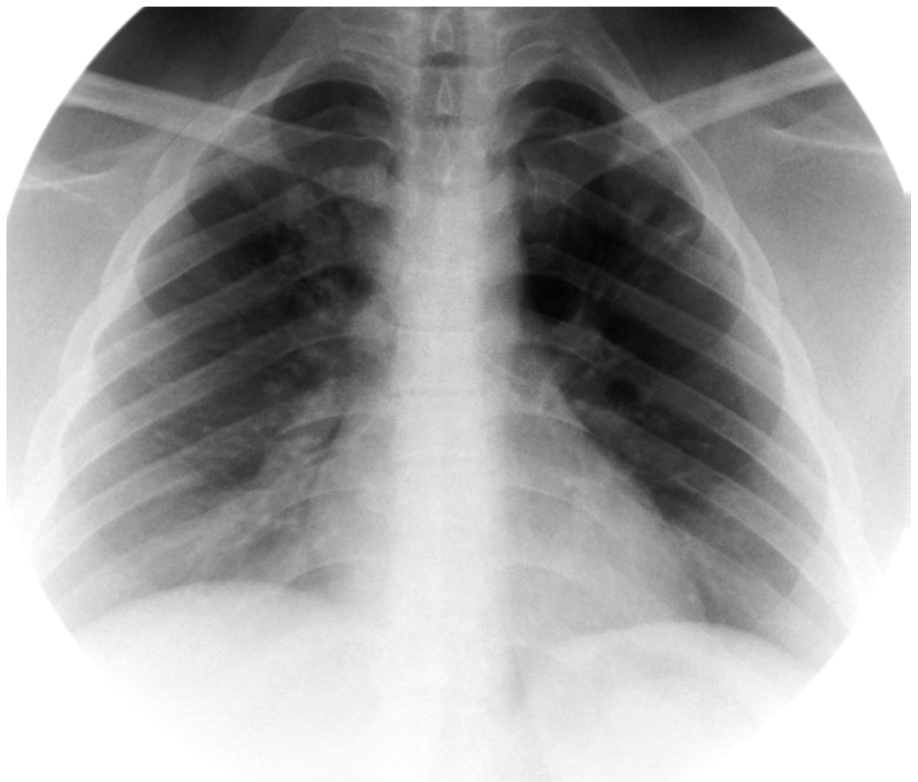
## Laser Resection

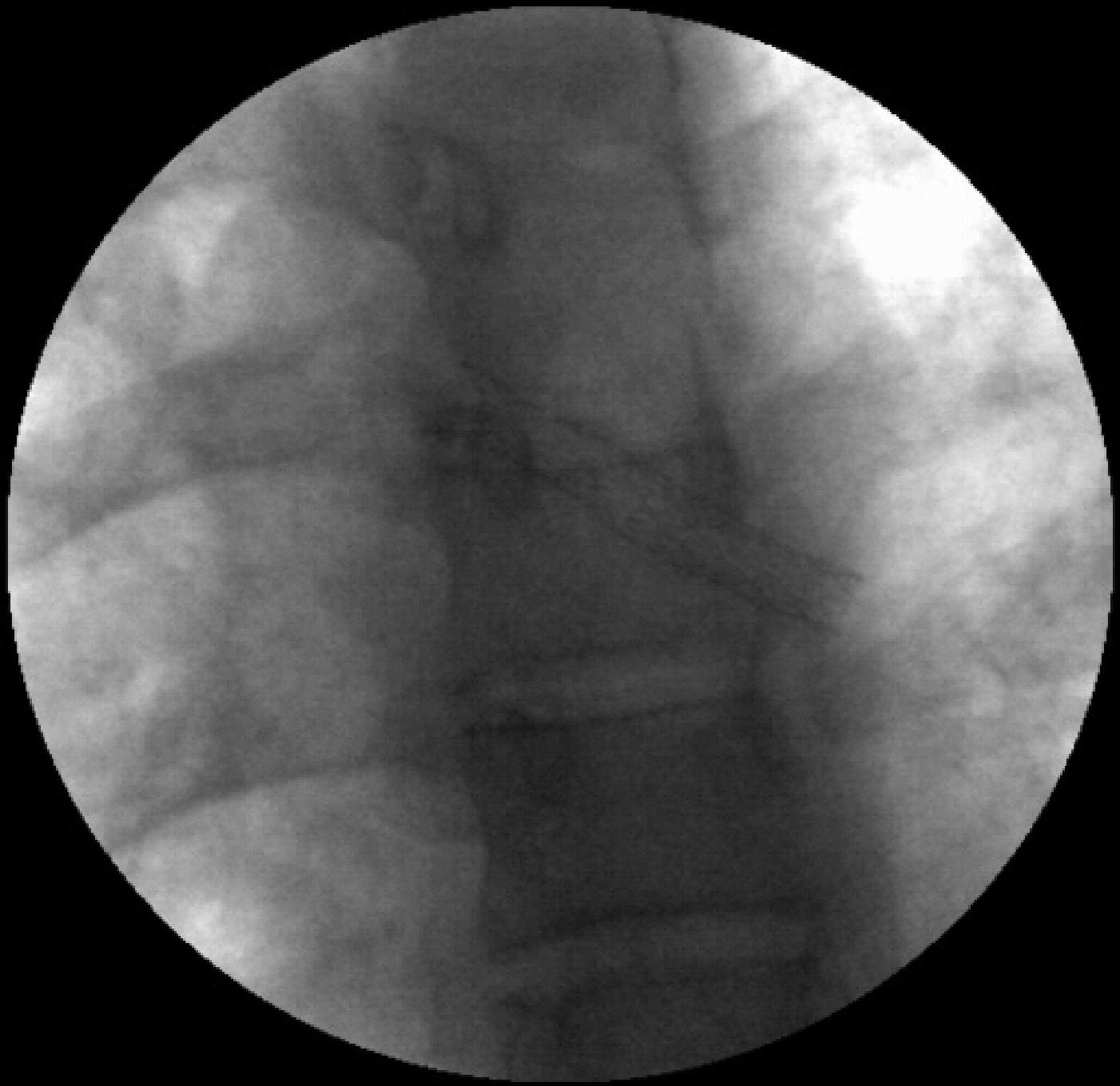
# Airway Stenting

## Cicatrical post-TB bronchial stenosis

A 24-year-old woman, after treatment with antituberculous medications for pulmonary tuberculosis, suffered by cough, dyspnea, wheezing. CT scan showed stricture of the left main bronchus starting from bifurcation to the end and having a lumen diameter 2mm. Bronchoscopic examination revealed focal narrowing to pinhole size to the left main bronchus with severe fibrotic changes. Under general anesthesia, balloon-expendable, metallic, non-covered stent (Medtronic) was inserted across the stenotic lesion and was expanded under the pressure of 9 atmospheres using Indeflator Perouse Medical. Bronchography, taken immediately after stent insertion, showed an expanded left main bronchus.



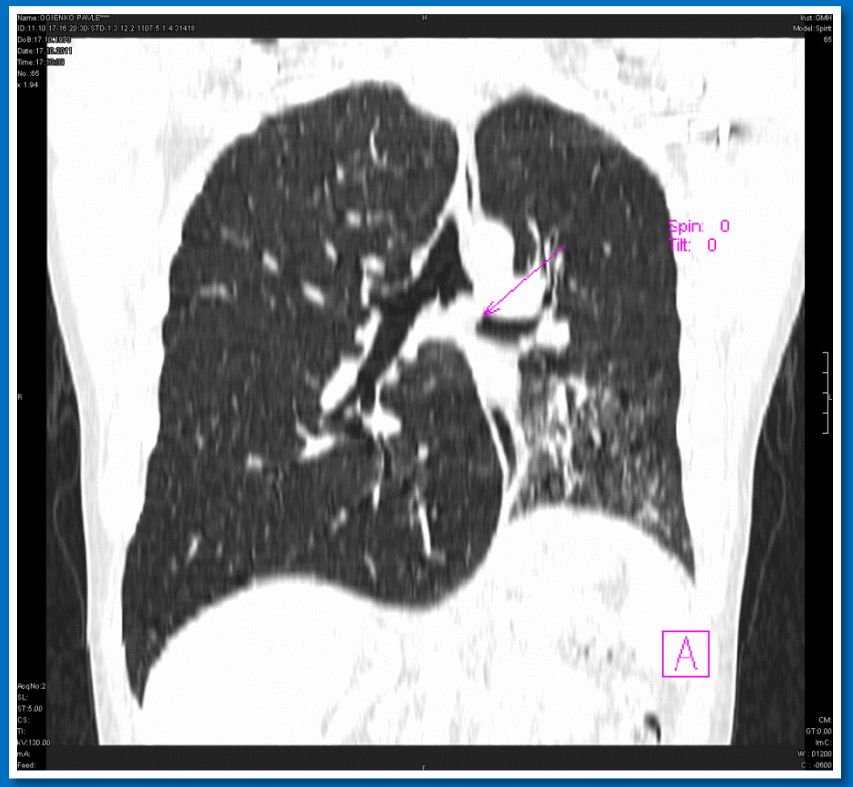


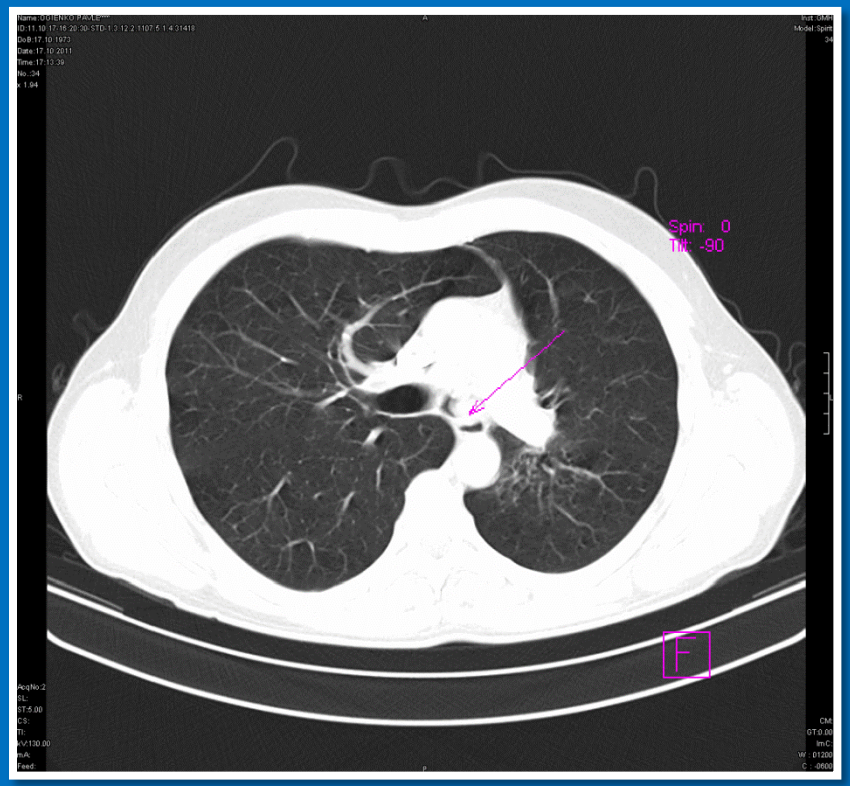


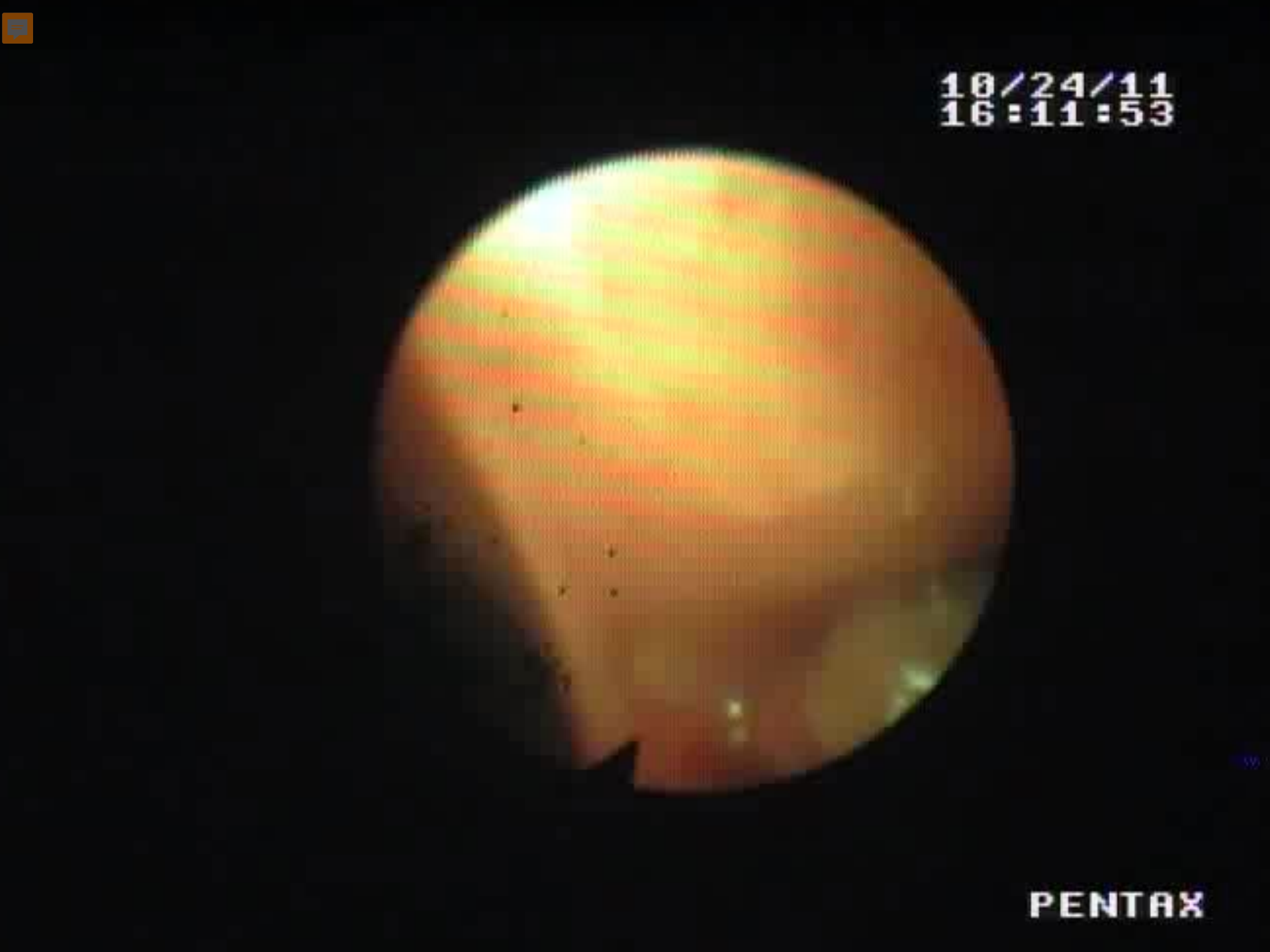
# Laser Resection

## Basal cell adenoma

**39 year old male. Bronchoscopically and by chest CT scanning was diagnosed with tumor of the left main bronchus, which is almost entirely strangled lumen. Biopsy materials cyto - morphological and molecular study revealed “Salivary gland like”-tumor. Surgeons refrained from surgery because of its location. The patient's condition worsened, and under vital indications was performed laser resection using VELASTM 30WTM Surgical Diode Laser System .**

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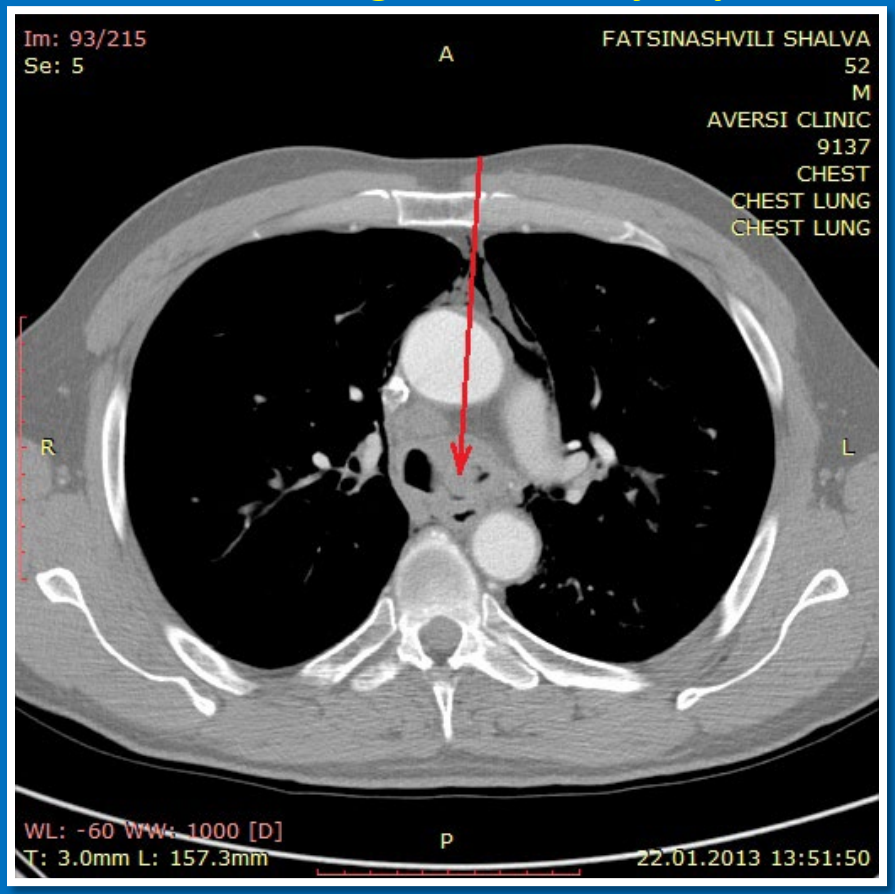
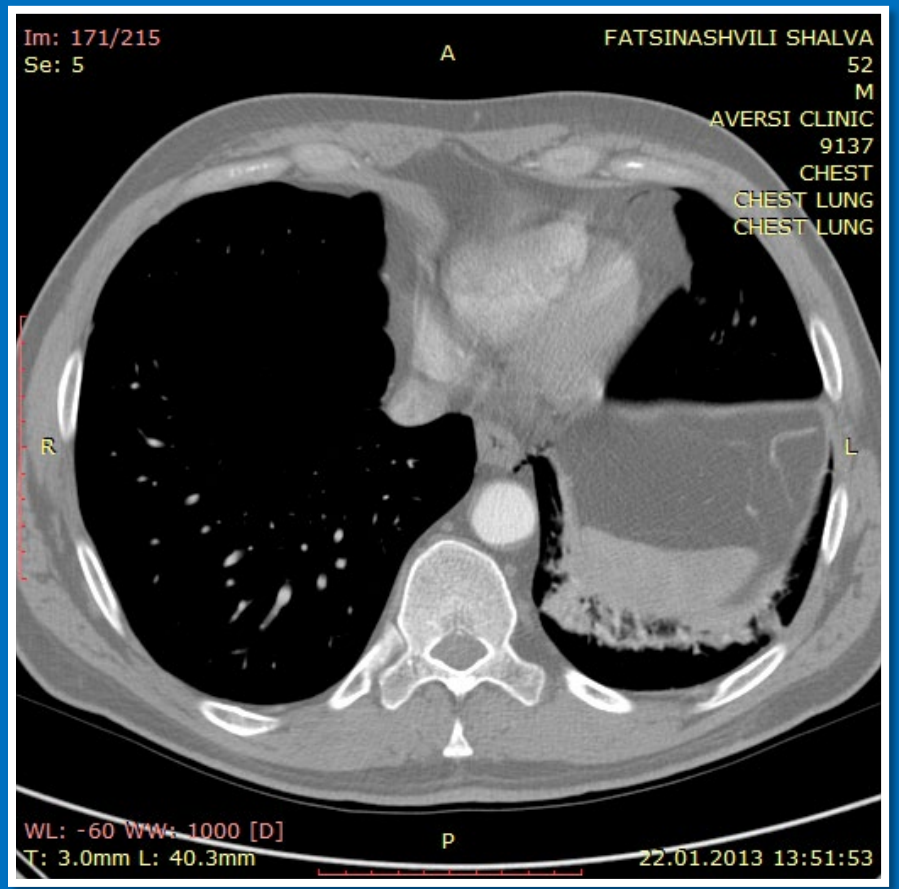
VELAS

# Laser Resection

## squamous-cell carcinoma

52 year old male. One month of medical history. Chest CT revealed subcarinal tissue mass that completely blocked the lumen of the left main bronchus and the right upper lobe bronchus. Endobronchial biopsy revealed squamous-cell carcinoma. The patient's condition worsened, He wasn’t able to sleep for  last 2 weeks due to suffocation and under vital indications the laser resection was performed using VELASTM 30WTM Surgical Diode Laser System. As a result, his breathing immediately improved . Now he is undergoing chemotherapy.



# THANK YOU FOR YOUR ATTENTION!