## 146P Clinical significance of ROS1 5′ deletions detected by FISH and response to crizotinib

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**Background:** ROS1-rearranged non-small cell lung cancer (NSCLC) patients (pts) are eligible for crizotinib therapy. Diagnosis is based on break-apart fluorescence in situ hybridization (FISH), as for ALKrearrangements, and include 5' deletions. We report assessment of ROS15' deletion by FISH and next-generation sequencing (NGS) and outcome of crizotinib treatment.

**Methods:** We included all consecutive NSCLC pts treated at our Institution with: i) diagnosis of 5' ROS1 gene fusion by break-apart FISH (≥15% of tumor cells with any 5'

**Table: 146P**

| Patient | Age | Stage at diagnosis | FISH break-apart status % of rearranged nuclei | | ROS1 gene fusion at NGS (partner gene) | | Line of crizotinib administration | Objective Response | Last follow-up (D=Dead A=Alive) | Survival after crizotinib, months | Still on treatment | Time on treatment, months |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| #1 | 52 | IVA | 5′ deletion | 41% | – | | II | PD | D | 4.5 | NO | 3.5 |
| #2 | 67 | IIIB | 5′ deletion | 60% | + | (EZR) | I | CR | A | 31.1 | YES | 31.1 |
| #3 | 60 | IVA | 5′ deletion | 52% | + | (EZR) | II | CR | A | 19.8 | YES | 19.8 |
| #4 | 54 | IVA | 5′ deletion | 44% | – | | II | PR | A | 15.7 | YES | 15.7 |
| #5 | 58 | IVA | 5′ deletion | 32% | - * | | IV | PR | A | 16.7 | YES | 16.7 |
| #6 | 53 | IVA | 5′ deletion | 30% | – | | II | PD | D | 2.1 | NO | 0.7 |
| #7 | 62 | IVA | 5′ deletion | 48% | + | (EZR) | II | PR | A | 6.4 | YES | 6.4 |
| #8 | 46 | IVB | 5′ deletion | 56% | + | (SCD4) | II | PR | A | 2.1 | NO | 2.1 |

*Case #5 NGS showed EML4-ALKrearrangement, subsequently confirmed by FISH.

deletion pattern); ii) availability in the samples of at least 50 ng of extracted RNA with at least 50% tumor cell enrichment; iii) treatment with crizotinib for at least 4 weeks following the diagnosis of ROS1fusion; iv) availability of clinical and radiological response data after therapy. FISH assay was performed using the Zytolight SPEC ROS1 Dual Color Break Apart Probe (ZytoVision, Germany). NGS was performed on Ion Torrent Personal Genome Machine (Thermo Fisher Scientific). The RNA panel identified rearrangements in 23 genes including ROS1 rearrangements with EZR, CD74 and SCD4.

**Results:** Eight patients were included. No patient had brain metastasis at diagnosis. Five pts were never-smoker, 2 light former smoker and 1 (case #1) a current heavy smoker (50 pack-year). Crizotinib therapy lasted for a mean of 11.0 months (range 2-31). The median overall survival was not reached at a median follow-up of 11.1 months (15.7 months for censored only). In 4 of the 8 cases (cases #2, 3, 7, 8; 50%), NGS confirmed a ROS1 fusion: 3 of them with the partner EZR and 1 with SCD4. All of these pts showed an objective response to crizotinib, 2 of them being complete responses according to RECIST v1.1 criteria. All these pts were alive at the time of last follow-up. In the other 4 pts (cases # 1, 4 ,5 ,6), NGS analysis did not detect ROS1 fusions. Of these, objective response to crizotinib was observed in only 2 pts, including one (case #5) with a concomitant EML4-ALKrearrangement, confirmed by FISH. The two other patients experienced rapid progressive disease.

**Conclusions:** FISH-detected ROS15' deletion is associated with a high response probability to crizotinib, similarly to classical ROS1 gene rearrangement. However, confirmation with at least one other method, e.g. NGS, is recommended, in order to exclude possible false positive results.

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