Background Even though immune checkpoint inhibitors have improved the survival of non-small cell lung cancer (NSCLC) patients, their prognosis is still poor. Natural Killer (NK) cells are lymphocytes with distinct anti-cancer properties, and NK cell activity (NKA) may have a prognostic role. Similarly, derived Neutrophils-to-lymphocyte ratio (dNLR) has been suggested as a prognostic biomarker in cancer patients.

Methods Patients with advanced or recurrent NSCLC eligible for anti-PD-1/PD-L1 treatment were enrolled at the Department of Oncology, Vejle Hospital, Denmark. Blood was sampled at baseline and immediately before the following three cycles of therapy. Interferon gamma (IFNγ) as a surrogate for NKA was measured using the NK Vue® assay (NKMAX, Seongnam-si, South Korea). Normal levels of IFNγ were considered as >250 pg/mL. Neutrophils and leukocytes were quantified by XN-9000 (Sysmex, Kobe, Japan). dNLR was calculated as (neutrophils/(leukocytes-neutrophils)) and dNLR<3 was considered normal. The study was approved by the Regional Committee on Health Research Ethics for Southern Denmark, approval number S-20170063.

Results Baseline level of NKA had no prognostic impact for overall survival (OS) (p=0.1279, NKA-normal n=45 vs NKA-low n=27, logrank). Longitudinal sampling allowed us to determine if dynamics in NKA were of prognostic value. For this purpose, patients were grouped as follows: NKA-low group had NKA <250 pg/mL at all available time-points (n=13), NKA-mixed group had NKA of varying levels (n=34), and NKA-high group had NKA >250 pg/mL at all available time-points (n=29). A logrank test revealed a significant difference between the groups in terms of OS (p=0.0002). Compared to the NKA-high group, the NKA-low group had a poor prognosis with a hazard ratio (HR) of 3.690 (p=0.001). After adjusting for dNLR, histology and performance status it remained an independent prognostic factor (HR=3.581, p=0.005). Interestingly, while dNLR also had prognostic impact for overall survival (OS) (p=0.0274, dNLR<3 n=57 and dNLR>3 n=18, logrank) and dNLR>3 was a prognostic marker for poor OS in univariate analysis (HR=1.908, p=0.03), this was lost in multivariate analysis taking NKA, histology, and performance status into account (HR=1.364, p=0.451).

Conclusions NKA dynamics, and not dNLR, was an independent prognostic marker for OS in NSCLC patients receiving anti-PD-1/PD-L1 treatment. These findings highlight the value of not restricting biomarker evaluation to immune cell enumeration, but rather looking at the ability of these cells to carry out vital immune functions.

Ethics Approval The study was approved by the Regional Committee on Health Research Ethics for Southern Denmark, approval number S-20170063. All participants provided written informed consent.