

## Review

---

# New Insights and Implications of Natural Killer Cells in Parkinson's Disease

Kelly B. Menees and Jae-Kyung Lee\*

*Department of Physiology and Pharmacology, University of Georgia College of Veterinary Medicine, Athens, GA, USA*

Accepted 12 April 2022  
Pre-press 9 May 2022

**Abstract.** Parkinson's disease (PD) is the second most common neurodegenerative disease and is characterized by the loss of dopaminergic neurons in the substantia nigra and the abnormal aggregation and accumulation of the alpha-synuclein ( $\alpha$ -syn) protein into Lewy bodies. It is established that there is an association between inflammation and PD; however, the time course of the inflammatory process as well as the immune cells involved are still debated. Natural killer (NK) cells are innate lymphocytes with numerous functions including targeting and killing infected or malignant cells, antimicrobial defense, and resolving inflammation. NK cell subsets differ in their effector function capacities which are modulated by activating and inhibitory receptors expressed at the cell surface. Alterations in NK cell numbers and receptor expression have been reported in PD patients. Recently, NK cell numbers and frequency were shown to be altered in the periphery and in the central nervous system in a preclinical mouse model of PD. Moreover, NK cells have recently been shown to internalize and degrade  $\alpha$ -syn aggregates and systemic NK cell depletion exacerbated synuclein pathology in a preclinical mouse model of PD, indicating a potential protective role of NK cells. Here, we review the inflammatory process in PD with a particular focus on alterations in NK cell numbers, phenotypes, and functions.

**Keywords:** Parkinson's disease, NK cells, immune system, inflammation

## INTRODUCTION

Parkinson's disease (PD) is the second most common neurodegenerative disease and is estimated to affect 1 million people and cost \$52 billion each year in the United States alone [1]. While some treatments for symptoms exist, there are currently no disease modifying treatments available for PD. PD is characterized by the abnormal aggregation of the alpha-synuclein ( $\alpha$ -syn) protein and dopaminergic degeneration in the substantia nigra *pars compacta* (SNpc). In recent years, the role of the immune

system and inflammation in PD has gained substantial ground. Elevated levels of pro-inflammatory cytokines including, interleukin (IL)-2, IL-6, and tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) have been reported in PD patient cerebrospinal fluid (CSF) [2]. Extracellular  $\alpha$ -syn aggregates have been found in the CSF and blood plasma of PD patients indicating the potential for  $\alpha$ -syn to modulate immune responses in the central nervous system (CNS) and periphery [3–6]. Recently, natural killer (NK) cells were shown to be scavengers of  $\alpha$ -syn aggregates and systemic depletion of NK cells in a preclinical mouse model of PD exacerbated synuclein pathology [7], thus implicating a potential protective role of NK cells in the context of PD. Investigating how immune cell populations, including NK cells, are altered throughout the course of PD may help indicate potential therapeutic

---

\*Correspondence to: Jae-Kyung Lee, PhD, Department of Physiology and Pharmacology, University of Georgia College of Veterinary Medicine, 501 D.W. Brooks Drive, Athens, GA 30602, USA. E-mail: jamlee@uga.edu.

targets. Here, we review the inflammatory processes in PD with a particular focus on NK cells. We highlight data from mouse models of PD as well as human peripheral blood mononuclear cell (PBMC) data that illustrate alterations in NK cell numbers, phenotypes, and functions. The effect of aging on NK cells and associated implications for PD are also discussed.

## NATURAL KILLER (NK) CELLS

NK cells are granular lymphocytes that develop from hematopoietic progenitors [8] and have been found to reside in lymphoid and non-lymphoid tissues [9]. In humans, NK cells comprise 10–15% of total circulating lymphocytes [10]. NK cells serve an important role in bridging the innate and adaptive immune systems through the secretion of cytokines and direct interaction with other immune cells [11]. NK cells are capable of targeting and destroying malignant or virus infected cells through direct lysis [12]. NK cells are able to destroy cells lacking major histocompatibility complex I (MHC I) (“missing-self”) without prior sensitization [13]. More diverse roles for NK cells have recently been described, including resolving inflammation [14, 15], modulating adaptive immunity [16, 17], antimicrobial defense [18, 19], clearance of senescent cells [20], and formation of immunological memory [21]. Effector functions of NK cells are modulated by signaling through a wide array of activating and inhibitory receptors (receptors/ligands of mouse and human NK cells reviewed in [22]). Alterations in NK cell receptors have been reported in PD patients (discussed below).

Human NK cells develop from cluster of differentiation (CD) 45+ common lymphoid progenitors (CLPs) which can also develop into B cells and T cells [23]. Human NK cells are distinguished from other lymphocytes by the expression of CD56 and CD16<sup>+/-</sup> and lack of CD3, which distinguishes NK cells from T cells [9]. CD markers are molecules expressed at the cell surface of immune cells and are used to identify leukocyte and lymphocyte subsets [24]. For an in-depth review of NK cell development see [9, 25]. Human NK cells are most commonly divided into two major subsets based on CD56 expression: CD56<sup>dim</sup> and CD56<sup>bright</sup> [26]. A third human NK cell subset, CD56<sup>-</sup>, has been reported and this population is increased in cases of chronic viral infection [27–29]. The major human NK cell subsets and their functions are summarized

Table 1  
Major human NK cell subsets

NK Cell Subset	Characteristics & Functions	References
CD56 <sup>bright</sup>	Robust cytokine production (IFN $\gamma$ , TNF $\alpha$ , GM-CSF, IL-10, IL-13)	[25, 30]
	Regulatory, immunomodulatory role	[25, 71]
	↓ with age	[85–87]
CD56 <sup>dim</sup>	Major subset in CSF	[34]
	High cytotoxic capacity	[30, 32]
	↑ perforin, granzymes, cytolytic granules	[31]
CD56 <sup>-</sup>	↑ with age	[87]
	Major population in circulation (>90%)	[25]
	↑ in chronic viral infections	[27–29]
	↓ cytotoxic capacity	[28]
	↓ cytokine production	[28]

↑: Increased; ↓: Decreased.

in Table 1. Up to 90% of circulating human NK cells are CD56<sup>dim</sup>CD16<sup>+</sup> [25]. CD56 can also be used to determine NK cell maturation status. CD56<sup>bright</sup> NK cells are considered to be immature while CD56<sup>dim</sup> NK cells are considered to be mature [30]. Compared to CD56<sup>bright</sup>CD16<sup>-</sup> NK cells, CD56<sup>dim</sup>CD16<sup>+</sup> NK cells have naturally higher cytotoxic effector functions, increased perforin, granzymes and cytolytic granules, and were shown to be the primary NK cell subset responsible for anti-tumor cytotoxicity [31, 32]. The CD56<sup>bright</sup>CD16<sup>-</sup> population is thought to serve a more regulatory role [25] and produce large amounts of pro-inflammatory cytokines such as IFN- $\gamma$ , TNF- $\alpha$ , IL-10, IL-13, and GM-CSF [33]. In human CSF samples, the majority of NK cells are CD56<sup>bright</sup> [34]. Murine NK cells are defined by their expression of CD11b and CD27 [9]. Expression of CD11b and CD27 change based on the maturation of murine NK cells and can be used to define maturation subsets: CD11b<sup>low</sup>CD27<sup>low</sup> (immature), CD11b<sup>low</sup>CD27<sup>high</sup> (intermediate), CD11b<sup>high</sup>CD27<sup>high</sup> (intermediate), to CD11b<sup>high</sup>CD27<sup>low</sup> (mature) [35]. CD27<sup>+</sup>CD11b<sup>+</sup> NK cells have increased effector functions [9].

## NK CELLS IN PD

NK cells have been identified as a biologically meaningful cluster of cells within the brain [36]. We recently demonstrated that NK cells internalize and degrade  $\alpha$ -syn aggregates *in vitro* through the endosomal/lysosomal pathway [7]. Immunohistochemistry analysis has shown the presence of NK

Table 2

Reported NK cell changes in PD patients compared to healthy controls

NK cell Population	↑/↓/NC Compared to HC	Reference
CD3 <sup>-</sup> CD56 <sup>+</sup>	↑	[41]
CD56 <sup>+</sup> or CD16 <sup>+</sup>	↑	[39]
CD3 <sup>+</sup> CD56 <sup>+</sup>	↑	[40]
CD3 <sup>-</sup> CD56 <sup>+</sup> CD16 <sup>+</sup>	↑	[38]
Not defined	↑	[42]
CD56 <sup>+</sup> CD16 <sup>+</sup> CD57 <sup>-</sup> CD28 <sup>-</sup>	↑	[43]
CD56 <sup>+</sup> CD16 <sup>+</sup> CD57 <sup>+</sup> CD28 <sup>-</sup>	↑	[43]
CD56 <sup>+</sup> CD16 <sup>+</sup> CD57 <sup>+</sup> CD28 <sup>+</sup>	↓	[43]
NK cell Receptors		
CD3 <sup>-</sup> CD56 <sup>+</sup> NKG2D <sup>+</sup>	NC	[41]
CD3 <sup>-</sup> CD56 <sup>+</sup> CD314 <sup>+</sup> (NKG2D)	↑	[39]
CD3 <sup>-</sup> CD56 <sup>+</sup> NKG2A <sup>+</sup>	↓	[41]

HC: healthy control; ↑: Increased; ↓: Decreased; NC: no change.

cells in postmortem synucleinopathy patient brain samples where robust p- $\alpha$ -syn inclusions were also located [7], indicating proximity to scavenge  $\alpha$ -syn aggregates. Importantly, in the presence of  $\alpha$ -syn aggregates NK cell cytotoxicity was attenuated and IFN- $\gamma$  secretion was decreased, indicating that NK cells were not aberrantly activated by  $\alpha$ -syn [7]. This contrasts the response microglia display to  $\alpha$ -syn aggregates which leads to the production of pro-inflammatory cytokines and reactive oxygen species [37]. Therefore, this newly identified function of NK cells may prove to be a useful therapeutic tool to reduce  $\alpha$ -syn burden without activation and potential increases in inflammation.

Several groups have reported higher percentages of NK cells in PD patient blood samples compared to healthy controls determined via flow cytometric analyses [38–41] (summarized in Table 2). It is important to note that most of the available data regarding NK cell alterations in human samples do not distinguish NK cell subsets but rather, group all NK cells as CD56<sup>+</sup> without regard for the distinct changes that may be occurring in the various NK cell populations. Huang et al. recently reported resting NK cells are significantly increased in PD patient peripheral blood compared to healthy controls using the CIBERSORT method [42]. Furthermore, utilizing gene set enrichment analysis (GSEA) of Kyoto encyclopedia of genes and genomes (KEGG) showed that NK cell mediated cytotoxic pathways were enriched in blood of PD patients [42], implicating potential increased activation status of NK cells within the periphery. Tian et al. reported an increase in frequency of CD56<sup>+</sup> CD16<sup>+</sup> CD57<sup>-</sup> CD28<sup>-</sup> and CD56<sup>+</sup> CD16<sup>+</sup> CD57<sup>+</sup> CD28<sup>-</sup> NK cell

populations in PD patients compared to healthy controls [43]. CD28 has been considered an activating receptor on NK cells and through its interaction with other receptors is associated with degranulation, target cell lysis, and production of proinflammatory cytokines [44]. CD57 expression is considered a marker of NK cell maturation and is associated with high cytotoxic capacity [45]. Tian and colleagues also demonstrated that increased CD57<sup>-</sup>CD28<sup>-</sup> NK cells were associated with increased Unified Parkinson's Disease Rating Scale (UPDRS) scores, indicating an increased population of less cytotoxic NK cells with disease severity [43]. Further analysis should be done to evaluate alterations in NK cell cytotoxic pathways at varying stages of PD, as increased or decreased cytotoxic activity may influence potential protective mechanisms of NK cells. It is generally accepted in the field that NK cell cytotoxicity declines with age [46], so evidence that NK cell mediated cytotoxic pathways are enriched in PD patient blood may indicate a potential immunotherapeutic target. While evidence of increased cytotoxic pathways has been reported, functional assays are needed to truly evaluate these results. Moreover, while cytotoxicity is a critical NK cell effector function, further evaluation of the role of NK cell cytotoxicity in PD is warranted to draw conclusions surrounding alterations in cytotoxic NK cell subsets. Alterations in NK cell receptors have also been reported. NK cell function is mediated by a variety of activating and inhibitory receptors expressed at the cell surface [47]. There are three major subsets of human NK cell receptors: killer cell Ig-like receptor (KIR) superfamily, C-type lectin superfamily, and natural cytotoxicity receptors (NCRs) [48]. KIRs are a family of activating and inhibitory receptors that are thought to be involved in the development of NK cell tolerance [49]. KIRs are encoded by the highly polymorphic *KIR* genes [50]. A recent study investigated the allelic variation of *KIR* in relation to PD symptoms and found KIR3DL1 variants protect from motor symptoms of PD [50]. NKG2A is an inhibitory receptor that recognizes HLA-E [51]. PD patients displayed decreased NKG2A<sup>+</sup> NK cells compared to non-PD controls [41]. Conflicting findings of NKG2D have been reported. Mihara et al. reported no difference in frequency of NKG2D<sup>+</sup> NK cells in PD patients compared to healthy controls [41], while Niwa et al. reported increased percentage of NKG2D<sup>+</sup> NK cells in PD patients [39].

It is reported that up to 35% of PD patients also have comorbid depression, thus a recent study by

Green et al. examined the levels of p11, a protein implicated in depression, in peripheral leukocyte populations [52]. p11 is a member of the S100 EF-hand protein family [53]. Previous work has demonstrated that in rodent models of PD, p11 expression is upregulated in dopaminergic neurons following L-DOPA treatment [54–56], implicating a potential role in PD. Interestingly, levels of p11 in NK cells isolated from peripheral blood of PD patients were shown to be positively associated with severity of PD as determined by UPDRS score [52]. Therefore, p11 levels in peripheral blood NK cells may be another marker to monitor PD progression.

Idiopathic rapid eye movement sleep behavior disorder (iRBD) has been identified as a prodromal marker for synucleinopathies [57]. A recent study reported an increase in the percentage of CD56<sup>dim</sup> NK cells (defined as TLR2<sup>-</sup>CD56<sup>dim</sup>) in iRBD patients compared to healthy controls [58]. No difference in the CD56<sup>bright</sup> NK cell (defined as TLR2<sup>-</sup>CD56<sup>bright</sup>) population was observed [58]. These findings suggest that NK cell alterations may be occurring early on in synucleinopathy pathogenesis.

Gastrointestinal disturbances are common non-motor symptoms of PD patients and can be present for years prior to the development of motor symptoms [59]. Patients with inflammatory bowel disease (IBD) are more likely to develop PD than non-IBD patients (reviewed in [60]). NK cells have been implicated in IBD pathogenesis [61, 62]. *KIR* polymorphisms have been linked to IBD risk [63]. Thus, NK cell alterations may be occurring in the prodromal phase of PD. Further research is needed to investigate alterations in the gastrointestinal NK cell populations and how potential changes in this population may correlate with PD development.

Preliminary data from our lab suggests that expression of NK cell activating and inhibitory receptors may be altered on NK cell subsets in PD patients. Our data suggests no differences in frequency of NK cells between PD patients and healthy controls, however, NK cell subsets were altered in PD patients when samples were stratified by UPDRS score and/or disease duration (unpublished observation). Furthermore, our preliminary data indicates that NK cell receptors, such as NKG2A, NKG2D or CX3CR1, may be altered on NK cell subsets in PD patients (unpublished observation). Further investigation into receptor expression on NK cell subsets is needed to fully elucidate potential alterations in NK cell subsets throughout the course of PD pathogenesis.

Additionally, factors such as genetic mutations, medication usage, and sex should be considered when analyzing such data to provide a more complete understanding of these immune cell alterations.

## NK CELLS IN ANIMAL MODELS OF PD

Recently, we illustrated that intrastriatal injection of preformed fibril (PFF)  $\alpha$ -syn in WT altered immune cell profiles in the brain and periphery [64]. Synuclein pathology was observed in multiple brain regions (primary motor cortex, striatum, SNpc, and hippocampus) and was detected in the small intestines of PFF  $\alpha$ -syn injected mice 5 months post-injection [64]. In PFF  $\alpha$ -syn injected mice, NK cell frequency in the CNS parenchyma was increased compared to control mice that received monomer  $\alpha$ -syn injection [64]. Increased frequencies of B cells, CD4<sup>+</sup> and CD8<sup>+</sup> T cells, and CD11b<sup>+</sup>CD45<sup>high</sup> infiltrated macrophages and a decreased frequency of CD11b<sup>+</sup>CD45<sup>low</sup> microglia were also observed in the CNS parenchyma of PFF  $\alpha$ -syn injected mice compared to monomer [64]. Furthermore, a single intrastriatal PFF  $\alpha$ -syn injection altered the peripheral immune cell profiles of these mice 5 months post-injection [64]. Total leukocyte numbers were increased in the inguinal lymph nodes and spleens but not in the blood of PFF  $\alpha$ -syn injected mice compared to monomer  $\alpha$ -syn injected mice 5 months post-injection [64]. NK cell numbers were increased in the spleens of PFF  $\alpha$ -syn injected mice compared to monomer  $\alpha$ -syn injected mice, and decreased frequency and numbers were observed in the inguinal lymph nodes and blood, respectively [64]. These results suggest that a CNS initiated pathology is a sufficient trigger to influence the peripheral immune response.

We recently demonstrated that systemic NK cell depletion in a preclinical mouse model of synucleinopathy resulted in increased p- $\alpha$ -syn pathology in the striatum, SNpc, and brainstem [7]. The M83 transgenic mouse line, which overexpresses the human A53T mutant  $\alpha$ -syn protein [65], was utilized in combination with intrastriatal injection of PFF  $\alpha$ -syn (or monomer  $\alpha$ -syn as the control) in combination with a systemic NK cell depletion strategy. NK cells were systemically depleted via a NK1.1 monoclonal antibody (mAb) delivered via intraperitoneal injection beginning 2 days prior to stereotaxic injection of  $\alpha$ -syn and every 5 days thereafter [7]. Importantly, to confirm NK cell depletion flow cytometric

analysis of the brain, spleen and inguinal lymph nodes was performed [7]. When serum cytokines were analyzed in NK cell depleted mice (without  $\alpha$ -syn injection) it was observed that IFN- $\gamma$  was significantly diminished [7]. As NK cells are major producers of IFN- $\gamma$  [66], this observation further validates the effectiveness of the monoclonal antibody depletion strategy. However, the depletion of IFN- $\gamma$  raises the question of its importance in PD pathogenesis. Are the effects observed due to the lack of NK cells themselves, or the IFN- $\gamma$  they secrete? IFN- $\gamma$  is an important immune modulator with a wide range of functions (reviewed in [67]). Blood plasma levels of IFN- $\gamma$  have been shown to be increased in PD patients [68]. Further investigation into the role of IFN- $\gamma$  itself as a potential mediator of PD pathogenesis is warranted. Astrogliosis and microgliosis were also evaluated in these mice via immunohistochemistry. PFF  $\alpha$ -syn injected NK cell depleted mice displayed significantly increased GFAP (astrocyte marker) immunoreactivity in the striatum and SNpc and significantly increased Iba-1 (microglia marker) immunoreactivity in the striatum, SNpc, and brainstem [7], indicating a heightened neuroinflammatory status. The NK cell depleted PFF  $\alpha$ -syn injected mice displayed progressively increased clasping task scores [7]. Furthermore, NK cell depleted PFF  $\alpha$ -syn injected mice developed significantly increased clinical symptom scores [7]. The data from this study illustrates a potential protective role of NK cells in the context of PD. Thus, it could be postulated that an increased NK cell population may be beneficial. As discussed previously, NK cells are increased in the blood of PD patients compared to healthy controls. It is possible that this increase is reflective of a compensatory mechanism aimed at mitigating disease pathogenesis. Further evaluation of NK cell alterations in PD patients (e.g., receptor expression, subsets) and their function in PD patients is warranted.

### **NK CELLS IN AGING: IMPLICATIONS FOR PD**

The primary risk factor for development of neurodegenerative diseases is aging [69]. It is well established that aging significantly impacts the immune system. Throughout the aging process the immune system undergoes a process of reorganizational and compensatory modulations termed immunosenescence [70]. Aging results in an increase in the number of NK cells [46], a redistribution of

NK cell subsets, and altered phenotype and functions [71]. An increase in the CD56<sup>dim</sup> NK cell population is observed with age, while the CD56<sup>bright</sup> subset decreases [71]. NK cell cytotoxicity is thought to decline with age [46]. Aging may also contribute to impaired crosstalk between the innate and adaptive immune systems [46]. Therefore, aging may impact potential protective effects of NK cells in the context of PD. Despite increased numbers of NK cells with age, alterations in their effector functions may prove to be detrimental for PD pathogenesis.

Our recent analysis of the splenic immune cell profiles of young (2-3 months) and aged (18-22 months) C57BL/6J mice revealed a significant age-dependent decrease in NK cells (numbers and frequency) [72]. Importantly, sex differences were investigated in this study as many neurodegenerative diseases, including PD, are sex-biased [73]. Further analysis of the NK cell population in these mice revealed an age dependent increase in expression of CD107a (degranulation marker) and NKG2D (activating receptor) in males, but no age-dependent alterations in these markers in NK cells isolated from females [72]. No alterations in the expression of CX3CR1 (chemotactic marker) or NKG2A (inhibitory marker) were observed across age or sex [72]. These results indicate that in aged males, NK cells may be in a more activated or primed state and may contribute to a more pro-inflammatory state. Furthermore, when treated with IL-2, age-dependent increases in IFN- $\gamma$  production were observed in both sexes [72], indicating a hyperreactive response with age. Alterations in IFN- $\gamma$  production may have wide reaching effects as IFN- $\gamma$  is an important immune modulator. IFN- $\gamma$  aids in Th1 development, contributes to macrophage function, increases immune cell trafficking to infection sites, and induces expression of major histocompatibility complex class I and II on antigen presenting cells (reviewed in [67]). As previously mentioned, in PD patients blood plasma levels of IFN- $\gamma$  have been shown to be increased [68]. It was also reported that age may impact NK cell capacity to handle  $\alpha$ -syn aggregates in WT mice [72]. NK cells isolated from spleens of aged C57BL/6J female mice showed decreased internalization of  $\alpha$ -syn aggregates compared to young female mice [72]. However, NK cells from aged male mice showed no difference in internalization or clearance of  $\alpha$ -syn aggregates compared to young male mice [72]. Alterations in the efficiency of internalization and clearance of  $\alpha$ -syn aggregates may lead to increased synuclein burden thereby potentially contributing to disease progression.

## NK CELLS IN OTHER NEURODEGENERATIVE DISEASES

Much of what is known about NK cells in the context of CNS diseases is derived from investigations of their role in experimental autoimmune encephalomyelitis (EAE) models of multiple sclerosis. Studies have shown that NK cells homing to the CNS was essential for ameliorating disease in EAE [74], implicating that NK cells exert protective effects. An immunoregulatory and neuroprotective role for NK cells has been suggested due to their ability to migrate to the inflamed CNS and inhibit activation of autoreactive T cells through killing of activated microglia [33]. It was recently shown that the gut microbiome licenses IFN- $\gamma$  expression in NK cells which promotes TRAIL expression in astrocytes that limit CNS inflammation [75]. Additionally, NK cells have been used as an immunotherapeutic for glioblastoma [76–78], illustrating their potential as a treatment strategy for CNS diseases.

In a recent study investigating the effects of NK cells in Alzheimer's disease (AD), depletion of NK cells was shown to enhance cognitive function and reduce neuroinflammation in the 3xTg mouse model of AD [79], implicating a deleterious effect of NK cells in the context of AD. Importantly, in this study levels of amyloid- $\beta$  were not altered following NK cell depletion [79]. The role of NK cells in AD is reviewed in [71]. Jin et al. demonstrated that systemic NK cell depletion in aged wild type mice improved spatial learning in the Morris water maze and improved hippocampal long-term potentiation [80], further supporting the notion that NK cells may be detrimental to cognitive function. These studies present contradictory evidence of NK cell depletion to a study utilizing a similar NK cell depletion strategy in a PD model [7], thus indicating that disease context may influence protective and/or negative effects of NK cells. However, it is also important to note that differences in NK cell depletion strategies may also influence outcomes. In the study from our group, M83 Tg mice (8–10 weeks old) received an initial dose of 100  $\mu$ g of anti-NK1.1 monoclonal antibody (mAb) 2 days prior to stereotaxic injection of  $\alpha$ -syn and thereafter 50  $\mu$ g of anti-NK1.1 mAb was administered every 5 days for 10 weeks, the entire duration of the study [7]. Zhang et al. administered 25  $\mu$ g of anti-NK1.1 mAb in 7–8 month old 3xTg-AD mice every 4 days for 4 weeks, the end of experiments [79], and Jin et al. administered 100  $\mu$ g of anti-NK1.1 mAb in aged WT mice (18

months old) every 5 days for 1 month, the end of experiments [80]. In addition to differences in disease/pathological contexts (PD, AD, aging) and NK cell depletion strategies, the various mouse lines utilized in these studies may also influence outcomes as differences in immune phenotypes and/or functions have been reported across different mouse strains in other disease contexts [81, 82].

Of importance, expression of ligands for NKG2D by damaged neurons has recently been reported. In a model of peripheral nerve injury, it was demonstrated that retinoic acid early transcript 1 (RAE1), a ligand for the NK cell activating receptor NKG2D, is expressed on damaged neurons, leading to NK cell mediated neurodegeneration [83]. This NK cell-mediated clearance of damaged neurons led to decreased mechanical hypersensitivity post injury [83]. In a mouse model of amyotrophic lateral sclerosis (ALS), spinal cord neurons expressed increased levels of the NKG2D ligand MULT1 but not RAE1 [84]. Thus, in the presence of  $\alpha$ -syn aggregates neuronal expression of NK cell ligands needs to be evaluated. In addition to scavenging extracellular  $\alpha$ -syn aggregates [7], if damaged neurons express NK cell ligands in the context of PD, NK cells may exert protective effects by specifically targeting those neurons, thereby preventing further propagation of  $\alpha$ -syn aggregates.

## CONCLUSIONS AND FUTURE DIRECTIONS

Evidence from different neurodegenerative diseases indicates there is not a “one-size-fits-all” when it comes to the effects of NK cells on disease phenotypes. The specific differences in pathologies and inflammatory states within different neurodegenerative diseases may influence the potential protective or deleterious effects of NK cells. The presence of NK cells in PD brains has the potential to impact and change the way we view the role of the immune response to  $\alpha$ -syn-induced inflammation and neurotoxicity in the CNS. The potential neuroprotective mechanisms of NK cells within the CNS include 1)  $\alpha$ -syn clearance, 2) changes in the expressions of ligands on neurons/glia for NK cell receptors and 3) interaction with infiltrated peripheral immune cells in the CNS.

Potential effects of NK cells in the periphery during synucleinopathy pathogenesis need to be further investigated, particularly within the gastrointestinal

system. Evidence has shown alterations in NK cell numbers and receptor expression in PD patients, but how these alterations influence disease pathogenesis/progression requires more investigation. Immune profiling studies to determine how different subsets of NK cells in the CNS and the periphery may be altered in PD will provide a novel direction to study potential mechanisms of NK cells in PD. In addition to the functional role of NK cells, NK cell immunosenescence associated with aging affects the health of older adults. Whether NK cell function/number diminishes with aging such that it results in the accumulation of  $\alpha$ -syn aggregates and decelerates resolution of immune responses will be an additional key question to be addressed.

Overall, the research surrounding the role of NK cells in PD is limited. While there is evidence that indicates that NK cells are altered in PD pathogenesis and can clear  $\alpha$ -syn aggregates *in vitro*, the exact role of NK cells in PD has yet to be fully elucidated. Furthermore, how NK cells potentially interact with neurons and glia in the CNS during PD pathogenesis needs to be explored. The mechanism behind NK cell internalization and degradation of  $\alpha$ -syn aggregates also warrants further investigation.

## ACKNOWLEDGMENTS

This work was supported by the National Institutes of Health (R01NS11960-01).

## CONFLICT OF INTEREST

The authors have no conflict of interest to report.

## REFERENCES

- [1] Marras C, Beck JC, Bower JH, Roberts E, Ritz B, Ross GW, Abbott RD, Savica R, Van Den Eeden SK, Willis AW, Tanner CM, Parkinson's Foundation P4 Group (2018) Prevalence of Parkinson's disease across North America. *NPJ Parkinsons Dis* **4**, 21.
- [2] Schröder JB, Pawlowski M, Meyer zu Hörste G, Gross CC, Wiendl H, Meuth SG, Ruck T, Warnecke T (2018) Immune cell activation in the cerebrospinal fluid of patients with Parkinson's disease. *Front Neurol* **9**, 1081.
- [3] El-Agnaf OM, Salem SA, Paleologou KE, Curran MD, Gibson MJ, Court JA, Schlossmacher MG, Allsop D (2006) Detection of oligomeric forms of alpha-synuclein protein in human plasma as a potential biomarker for Parkinson's disease. *FASEB J* **20**, 419-425.
- [4] Lee PH, Lee G, Park HJ, Bang OY, Joo IS, Huh K (2006) The plasma alpha-synuclein levels in patients with Parkinson's disease and multiple system atrophy. *J Neural Transm (Vienna)* **113**, 1435-1439.
- [5] Mollenhauer B, Locascio JJ, Schulz-Schaeffer W, Sixel-Döring F, Trenkwalder C, Schlossmacher MG (2011)  $\alpha$ -Synuclein and tau concentrations in cerebrospinal fluid of patients presenting with parkinsonism: A cohort study. *Lancet Neurol* **10**, 230-240.
- [6] Tokuda T, Qureshi MM, Ardah MT, Varghese S, Shehab SA, Kasai T, Ishigami N, Tamaoka A, Nakagawa M, El-Agnaf OM (2010) Detection of elevated levels of  $\alpha$ -synuclein oligomers in CSF from patients with Parkinson disease. *Neurology* **75**, 1766-1772.
- [7] Earls RH, Menees KB, Chung J, Gutekunst CA, Lee HJ, Hazim MG, Rada B, Wood LB, Lee JK (2020) NK cells clear alpha-synuclein and the depletion of NK cells exacerbates synuclein pathology in a mouse model of alpha-synucleinopathy. *Proc Natl Acad Sci U S A* **117**, 1762-1771.
- [8] Cichocki F, Grzywacz B, Miller JS (2019) Human NK cell development: One road or many? *Front Immunol* **10**, 2078.
- [9] Abel AM, Yang C, Thakar MS, Malarkannan S (2018) Natural killer cells: Development, maturation, and clinical utilization. *Front Immunol* **9**, 1869.
- [10] Hu W, Wang G, Huang D, Sui M, Xu Y (2019) Cancer immunotherapy based on natural killer cells: Current progress and new opportunities. *Front Immunol* **10**, 1205.
- [11] Inngjerdingen M, Kveberg L, Naper C, Vaage JT (2011) Natural killer cell subsets in man and rodents. *Tissue Antigens* **78**, 81-88.
- [12] Zhang Y, Huang B (2017) The development and diversity of ILCs, NK cells and their relevance in health and diseases. *Adv Exp Med Biol* **1024**, 225-244.
- [13] Camous X, Pera A, Solana R, Larbi A (2012) NK cells in healthy aging and age-associated diseases. *J Biomed Biotechnol* **2012**, 195956.
- [14] Thoren FB, Riise RE, Ousback J, Della Chiesa M, Alsterholm M, Marcenaro E, Pesce S, Prato C, Cantoni C, Bylund J, Moretta L, Moretta A (2012) Human NK cells induce neutrophil apoptosis via an NKp46- and Fas-dependent mechanism. *J Immunol* **188**, 1668-1674.
- [15] Waggoner SN, Kumar V (2012) Evolving role of 2B4/CD244 in T and NK cell responses during virus infection. *Front Immunol* **3**, 377.
- [16] Martin-Fontecha A, Thomsen LL, Brett S, Gerard C, Lipp M, Lanzavecchia A, Sallusto F (2004) Induced recruitment of NK cells to lymph nodes provides IFN-gamma for T(H)1 priming. *Nat Immunol* **5**, 1260-1265.
- [17] Vitale M, Della Chiesa M, Carlomagno S, Pende D, Arico M, Moretta L, Moretta A (2005) NK-dependent DC maturation is mediated by TNFalpha and IFNgamma released upon engagement of the NKp30 triggering receptor. *Blood* **106**, 566-571.
- [18] Schmidt RL, Filak HC, Lemon JD, Potter TA, Lenz LL (2011) A LysM and SH3-domain containing region of the *Listeria monocytogenes* p60 protein stimulates accessory cells to promote activation of host NK cells. *PLoS Pathog* **7**, e1002368.
- [19] Small CL, McCormick S, Gill N, Kugathasan K, Santosuosso M, Donaldson N, Heinrichs DE, Ashkar A, Xing Z (2008) NK cells play a critical protective role in host defense against acute extracellular *Staphylococcus aureus* bacterial infection in the lung. *J Immunol* **180**, 5558-5568.
- [20] Sagiv A, Biran A, Yon M, Simon J, Lowe SW, Krizhanovsky V (2013) Granule exocytosis mediates immune surveillance of senescent cells. *Oncogene* **32**, 1971-1977.
- [21] Nikzad R, Angelo LS, Aviles-Padilla K, Le DT, Singh VK, Bimler L, Vukmanovic-Stejic M, Vendrame E, Ranganath T, Simpson L, Haigwood NL, Blish CA, Akbar AN, Paust S

- (2019) Human natural killer cells mediate adaptive immunity to viral antigens. *Sci Immunol* **4**, eaat8116.
- [22] Earls RH, Lee JK (2020) The role of natural killer cells in Parkinson's disease. *Exp Mol Med* **52**, 1517-1525.
- [23] Scoville SD, Freud AG, Caligiuri MA (2019) Cellular pathways in the development of human and murine innate lymphoid cells. *Curr Opin Immunol* **56**, 100-106.
- [24] Kalina T, Fišer K, Pérez-Andrés M, Kuzílková D, Cuenca M, Bartol SJW, Blanco E, Engel P, van Zelm MC (2019) CD maps—dynamic profiling of CD1–CD100 surface expression on human leukocyte and lymphocyte subsets. *Front Immunol* **10**, 2434.
- [25] Di Vito C, Mikulak J, Mavilio D (2019) On the way to become a natural killer cell. *Front Immunol* **10**, 1812.
- [26] Vivier E, Tomasello E, Baratin M, Walzer T, Ugolini S (2008) Functions of natural killer cells. *Nat Immunol* **9**, 503-510.
- [27] Mavilio D, Lombardo G, Benjamin J, Kim D, Follman D, Marcenaro E, O'Shea MA, Kinter A, Kovacs C, Moretta A (2005) Characterization of CD56–/CD16+natural killer (NK) cells: A highly dysfunctional NK subset expanded in HIV-infected viremic individuals. *Proc Natl Acad Sci U S A* **102**, 2886-2891.
- [28] Müller-Durovic B, Grählert J, Devine OP, Akbar AN, Hess C (2019) CD56-negative NK cells with impaired effector function expand in CMV and EBV co-infected healthy donors with age. *Aging* **11**, 724-740.
- [29] Tarazona R, Casado JG, Delarosa O, Torre-Cisneros J, Villanueva JL, Sanchez B, Galiani MD, Gonzalez R, Solana R, Pena J (2002) Selective depletion of CD56dim NK cell subsets and maintenance of CD56bright NK cells in treatment-naïve HIV-1-seropositive individuals. *J Clin Immunol* **22**, 176-183.
- [30] Poli A, Michel T, Thérésine M, Andrès E, Hentges F, Zimmer J (2009) CD56bright natural killer (NK) cells: An important NK cell subset. *Immunology* **126**, 458-465.
- [31] Jacobs R, Hintzen G, Kemper A, Beul K, Kempf S, Behrens G, Sykora KW, Schmidt RE (2001) CD56bright cells differ in their KIR repertoire and cytotoxic features from CD56dim NK cells. *Eur J Immunol* **31**, 3121-3127.
- [32] Penack O, Gentilini C, Fischer L, Asemissen AM, Scheibenbogen C, Thiel E, Uharek L (2005) CD56dimCD16neg cells are responsible for natural cytotoxicity against tumor targets. *Leukemia* **19**, 835-840.
- [33] Zhang C, Tian Z (2017) NK cell subsets in autoimmune diseases. *J Autoimmun* **83**, 22-30.
- [34] Han S, Lin YC, Wu T, Salgado AD, Mexhitaj I, Wuest SC, Romm E, Ohayon J, Goldbach-Mansky R, Vanderver A, Marques A, Toro C, Williamson P, Cortese I, Bielekova B (2014) Comprehensive immunophenotyping of cerebrospinal fluid cells in patients with neuroimmunological diseases. *J Immunol* **192**, 2551-2563.
- [35] Chiossone L, Chaix J, Fuseri N, Roth C, Vivier E, Walzer T (2009) Maturation of mouse NK cells is a 4-stage developmental program. *Blood* **113**, 5488-5496.
- [36] Van Hove H, Martens L, Scheyltjens I, De Vlaminck K, Pombo Antunes AR, De Prijck S, Vandamme N, De Schepper S, Van Isterdael G, Scott CL, Aerts J, Berx G, Boeckxstaens GE, Vandenbroucke RE, Vereecke L, Moechars D, Williams M, Van Ginderachter JA, Saey Y, Movahedi K (2019) A single-cell atlas of mouse brain macrophages reveals unique transcriptional identities shaped by ontogeny and tissue environment. *Nat Neurosci* **22**, 1021-1035.
- [37] Zhang QS, Heng Y, Yuan YH, Chen NH (2017) Pathological  $\alpha$ -synuclein exacerbates the progression of Parkinson's disease through microglial activation. *Toxicol Lett* **265**, 30-37.
- [38] Sun C, Zhao Z, Yu W, Mo M, Song C, Si Y, Liu Y (2019) Abnormal subpopulations of peripheral blood lymphocytes are involved in Parkinson's disease. *Ann Trans Med* **7**, 637.
- [39] Niwa F, Kuriyama N, Nakagawa M, Imanishi J (2012) Effects of peripheral lymphocyte subpopulations and the clinical correlation with Parkinson's disease. *Geriatr Gerontol Int* **12**, 102-107.
- [40] Cen L, Yang C, Huang S, Zhou M, Tang X, Li K, Guo W, Wu Z, Mo M, Xiao Y, Chen X, Yang X, Huang Q, Chen C, Qu S, Xu P (2017) Peripheral lymphocyte subsets as a marker of Parkinson's disease in a Chinese population. *Neurosci Bull* **33**, 493-500.
- [41] Mihara T, Nakashima M, Kuroiwa A, Akitake Y, Ono K, Hosokawa M, Yamada T, Takahashi M (2008) Natural killer cells of Parkinson's disease patients are set up for activation: A possible role for innate immunity in the pathogenesis of this disease. *Parkinsonism Relat Disord* **14**, 46-51.
- [42] Huang Y, Liu H, Hu J, Han C, Zhong Z, Luo W, Zhang Y, Ling F (2021) Significant difference of immune cell fractions and their correlations with differential expression genes in Parkinson's disease. *Front Aging Neurosci* **13**, 686066.
- [43] Tian J, Dai S-B, Jiang S-S, Yang W-Y, Yan Y-Q, Lin Z-H, Dong J-X, Liu Y, Zheng R, Chen Y, Zhang B-R, Pu J-L (2022) Specific immune status in Parkinson's disease at different ages of onset. *NPJ Parkinsons Dis* **8**, 5.
- [44] Zhuang X, Long EO (2019) CD28 homolog is a strong activator of natural killer cells for lysis of B7H7+tumor cells. *Cancer Immunol Res* **7**, 939-951.
- [45] Nielsen CM, White MJ, Goodier MR, Riley EM (2013) Functional significance of CD57 expression on human NK cells and relevance to disease. *Front Immunol* **4**, 422-422.
- [46] Hazeldine J, Lord JM (2013) The impact of ageing on natural killer cell function and potential consequences for health in older adults. *Ageing Res Rev* **12**, 1069-1078.
- [47] Vivier E, Raulet DH, Moretta A, Caligiuri MA, Zitvogel L, Lanier LL, Yokoyama WM, Ugolini S (2011) Innate or adaptive immunity? The example of natural killer cells. *Science* **331**, 44-49.
- [48] Cooper MA, Fehniger TA, Caligiuri MA (2001) The biology of human natural killer-cell subsets. *Trends Immunol* **22**, 633-640.
- [49] Pegram HJ, Andrews DM, Smyth MJ, Darcy PK, Kershaw MH (2011) Activating and inhibitory receptors of natural killer cells. *Immunol Cell Biol* **89**, 216-224.
- [50] Anderson KM, Augusto DG, Dandekar R, Shams H, Zhao C, Yusufali T, Montero-Martín G, Marin WM, Nemat-Gorgani N, Creary LE, Caillier S, Mofrad MRK, Parham P, Fernández-Viña M, Oksenberg JR, Norman PJ, Hollenbach JA (2020) Killer cell immunoglobulin-like receptor variants are associated with protection from symptoms associated with more severe course in Parkinson disease. *J Immunol* **205**, 1323-1330.
- [51] Sivori S, Vacca P, Del Zotto G, Munari E, Mingari MC, Moretta L (2019) Human NK cells: Surface receptors, inhibitory checkpoints, and translational applications. *Cell Mol Immunol* **16**, 430-441.
- [52] Green H, Zhang X, Tiklova K, Volakakis N, Brodin L, Berg L, Greengard P, Perlmann T, Svenningsson P (2017) Alterations of p11 in brain tissue and peripheral blood leukocytes

- in Parkinson's disease. *Proc Natl Acad Sci U S A* **114**, 2735-2740.
- [53] Gerke V, Weber K (1985) The regulatory chain in the p36-kd substrate complex of viral tyrosine-specific protein kinases is related in sequence to the S-100 protein of glial cells. *EMBO J* **4**, 2917-2920.
- [54] Zhang X, Andren PE, Greengard P, Svenningsson P (2008) Evidence for a role of the 5-HT<sub>1B</sub> receptor and its adaptor protein, p11, in L-DOPA treatment of an animal model of Parkinsonism. *Proc Natl Acad Sci U S A* **105**, 2163-2168.
- [55] Marongiu R, Arango-Lievano M, Francardo V, Morgestern P, Zhang X, Cenci MA, Svenningsson P, Greengard P, Kaplitt MG (2016) Gene therapy blockade of dorsal striatal p11 improves motor function and dyskinesia in parkinsonian mice. *Proc Natl Acad Sci U S A* **113**, 1423-1428.
- [56] Schintu N, Zhang X, Alvarsson A, Marongiu R, Kaplitt MG, Greengard P, Svenningsson P (2016) p11 modulates L-DOPA therapeutic effects and dyskinesia via distinct cell types in experimental Parkinsonism. *Proc Natl Acad Sci U S A* **113**, 1429-1434.
- [57] Dauvilliers Y, Schenck CH, Postuma RB, Iranzo A, Luppi P-H, Plazzi G, Montplaisir J, Boeve B (2018) REM sleep behaviour disorder. *Nat Rev Dis Primers* **4**, 19.
- [58] Farren K, Nissen SK, Stokholm MG, Iranzo A, Østergaard K, Serradell M, Otto M, Svendsen KB, Garrido A, Vilas D, Borghammer P, Santamaria J, Møller A, Gaig C, Brooks DJ, Tolosa E, Pavese N, Romero-Ramos M (2021) Monocyte markers correlate with immune and neuronal brain changes in REM sleep behavior disorder. *Proc Natl Acad Sci U S A* **118**, e2020858118.
- [59] Liddle RA (2018) Parkinson's disease from the gut. *Brain Res* **1693**, 201-206.
- [60] Brudek T (2019) Inflammatory bowel diseases and Parkinson's disease. *J Parkinsons Dis* **9**, S331-S344.
- [61] Yadav PK, Chen C, Liu Z (2011) Potential role of NK cells in the pathogenesis of inflammatory bowel disease. *J Biomed Biotechnol* **2011**, 348530.
- [62] Poggi A, Benelli R, Venè R, Costa D, Ferrari N, Tosetti F, Zocchi MR (2019) Human gut-associated natural killer cells in health and disease. *Front Immunol* **10**, 961.
- [63] Fathollahi A, Aslani S, Mostafaei S, Rezaei N, Mahmoudi M (2018) The role of killer-cell immunoglobulin-like receptor (KIR) genes in susceptibility to inflammatory bowel disease: Systematic review and meta-analysis. *Inflammation Res* **67**, 727-736.
- [64] Earls RH, Menees KB, Chung J, Barber J, Gutekunst CA, Hazim MG, Lee JK (2019) Intrastriatal injection of preformed alpha-synuclein fibrils alters central and peripheral immune cell profiles in non-transgenic mice. *J Neuroinflammation* **16**, 250.
- [65] Giasson BI, Duda JE, Quinn SM, Zhang B, Trojanowski JQ, Lee VM (2002) Neuronal alpha-synucleinopathy with severe movement disorder in mice expressing A53T human alpha-synuclein. *Neuron* **34**, 521-533.
- [66] Schoenborn JR, Wilson CB (2007) Regulation of interferon-gamma during innate and adaptive immune responses. *Adv Immunol* **96**, 41-101.
- [67] Mah AY, Cooper MA (2016) Metabolic regulation of natural killer cell IFN- $\gamma$  production. *Crit Rev Immunol* **36**, 131-147.
- [68] Mount MP, Lira A, Grimes D, Smith PD, Faucher S, Slack R, Anisman H, Hayley S, Park DS (2007) Involvement of interferon-gamma in microglial-mediated loss of dopaminergic neurons. *J Neurosci* **27**, 3328-3337.
- [69] Lopez-Otin C, Blasco MA, Partridge L, Serrano M, Kroemer G (2013) The hallmarks of aging. *Cell* **153**, 1194-1217.
- [70] Pinchuk LM, Filipov NM (2008) Differential effects of age on circulating and splenic leukocyte populations in C57BL/6 and BALB/c male mice. *Immun Ageing* **5**, 1.
- [71] Solana C, Tarazona R, Solana R (2018) Immunosenescence of natural killer cells, inflammation, and Alzheimer's disease. *Int J Alzheimers Dis* **2018**, 3128758.
- [72] Menees KB, Earls RH, Chung J, Jernigan J, Filipov NM, Carpenter JM, Lee JK (2021) Sex- and age-dependent alterations of splenic immune cell profile and NK cell phenotypes and function in C57BL/6J mice. *Immun Ageing* **18**, 3.
- [73] Gold SM, Willing A, Leyboldt F, Paul F, Friese MA (2019) Sex differences in autoimmune disorders of the central nervous system. *Semin Immunopathol* **41**, 177-188.
- [74] Hao J, Liu R, Piao W, Zhou Q, Vollmer TL, Campagnolo DI, Xiang R, La Cava A, Van Kaer L, Shi FD (2010) Central nervous system (CNS)-resident natural killer cells suppress Th17 responses and CNS autoimmune pathology. *J Exp Med* **207**, 1907-1921.
- [75] Sanmarco LM, Wheeler MA, Gutierrez-Vazquez C, Polonio CM, Linnerbauer M, Pinho-Ribeiro FA, Li Z, Giovannoni F, Batterman KV, Scalisi G, Zandee SEJ, Heck ES, Alsuwailm M, Rosene DL, Becher B, Chiu IM, Prat A, Quintana FJ (2021) Gut-licensed IFN $\gamma$ (+) NK cells drive LAMP1(+)/TRAIL(+) anti-inflammatory astrocytes. *Nature* **590**, 473-479.
- [76] Genbler S, Burger MC, Zhang C, Oelsner S, Mildenerger I, Wagner M, Steinbach JP, Wels WS (2016) Dual targeting of glioblastoma with chimeric antigen receptor-engineered natural killer cells overcomes heterogeneity of target antigen expression and enhances antitumor activity and survival. *Oncoimmunology* **5**, e1119354.
- [77] Han J, Chu J, Keung Chan W, Zhang J, Wang Y, Cohen JB, Victor A, Meisen WH, Kim SH, Grandi P, Wang QE, He X, Nakano I, Chiocca EA, Glorioso Iii JC, Kaur B, Caligiuri MA, Yu J (2015) CAR-engineered NK cells targeting wild-type EGFR and EGFRvIII enhance killing of glioblastoma and patient-derived glioblastoma stem cells. *Sci Rep* **5**, 11483.
- [78] Zhang C, Burger MC, Jennewein L, Genbler S, Schönfeld K, Zeiner P, Hattingen E, Harter PN, Mittelbronn M, Tonn T, Steinbach JP, Wels WS (2016) ErbB2/HER2-specific NK cells for targeted therapy of glioblastoma. *J Natl Cancer Inst* **108**, doi: 10.1093/jnci/djv375
- [79] Zhang Y, Fung ITH, Sankar P, Chen X, Robison LS, Ye L, D'Souza SS, Salinero AE, Kuentzel ML, Chittur SV, Zhang W, Zuloaga KL, Yang Q (2020) Depletion of NK cells improves cognitive function in the Alzheimer disease mouse model. *J Immunol* **205**, 502-510.
- [80] Jin WN, Shi K, He W, Sun JH, Van Kaer L, Shi FD, Liu Q (2021) Neuroblast senescence in the aged brain augments natural killer cell cytotoxicity leading to impaired neurogenesis and cognition. *Nat Neurosci* **24**, 61-73.
- [81] Sellers RS, Clifford CB, Treuting PM, Brayton C (2011) Immunological variation between inbred laboratory mouse strains: Points to consider in phenotyping genetically immunomodified mice. *Vet Pathol* **49**, 32-43.
- [82] Becker KJ (2016) Strain-related differences in the immune response: Relevance to human stroke. *Transl Stroke Res* **7**, 303-312.
- [83] Davies AJ, Kim HW, Gonzalez-Cano R, Choi J, Back SK, Roh SE, Johnson E, Gabriac M, Kim M-S, Lee J, Lee JE, Kim YS, Bae YC, Kim SJ, Lee K-M, Na HS, Riva P, Latremoliere A, Rinaldi S, Ugolini S, Costigan M, Oh SB (2019) Natural killer cells degenerate

- intact sensory afferents following nerve injury. *Cell* **176**, 716-728.e718.
- [84] Garofalo S, Coccozza G, Porzia A, Inghilleri M, Raspa M, Scavizzi F, Aronica E, Bernardini G, Peng L, Ransohoff RM, Santoni A, Limatola C (2020) Natural killer cells modulate motor neuron-immune cell cross talk in models of Amyotrophic Lateral Sclerosis. *Nat Commun* **11**, 1773.
- [85] Solana R, Campos C, Pera A, Tarazona R (2014) Shaping of NK cell subsets by aging. *Curr Opin Immunol* **29**, 56-61.
- [86] Chidrawar SM, Khan N, Chan Y, Nayak L, Moss PA (2006) Ageing is associated with a decline in peripheral blood CD56bright NK cells. *Immun Ageing* **3**, 1-8.
- [87] Le Garff-Tavernier M, Béziat V, Decocq J, Siguret V, Gandjbakhch F, Pautas E, Debré P, Merle-Beral H, Vieillard V (2010) Human NK cells display major phenotypic and functional changes over the life span. *Aging Cell* **9**, 527-535.