# Title: An ethnomedical toolkit to discover plant uses of therapeutic potential for neurodegenerative diseases


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**ABSTRACT**

**Background:** Neurodegenerative diseases (NDs) are mostly ultimately fatal, with pharmacological interventions failing to halt or reverse disease progression. These disorders include NDs of childhood such as Batten disease. 50 % of therapeutic drugs are derived from natural products including plants. However, the majority of over 350,000 known plant species await investigation for biological activity, representing a huge untapped resource.

**Methods:** This study employed a novel toolkit methodology, in which therapeutic categories of ND relevance were translated into terms used in ethnomedicine. The literature was searched using electronic databases and relevant articles. This enabled an assessment of ND pathologies and symptoms, in order to construct therapeutic categories recognized by ethnomedical practitioners. These categories were then applied to find plant species with reported therapeutic effects of ND relevance, mined from 157 ethnomedical surveys and 2 databases. The findings were analysed in their ecological contexts, to determine rich locations of ethnomedical data, and how this data may be at risk.

**Results:** 2001 plant species were identified with reported uses for alleviating pathologies relevant to NDs. Uses were documented for therapeutic categories such as memory improvement (reported in 130 species), anti-paralytic (185 species), anti-toxin (125 species) and other beneficial effects. Species were found with therapeutic potential across numerous NDs ranging from Alzheimer's disease to rare NDs such as motor neurone disease and Friedreich's ataxia. 734 species demonstrated multiple therapeutic uses. There is a trend of relatively more remedies for serious diseases where ethnomedical plant use is vital to the community. 57 % of the studies were found to reside within biodiversity hotspots (defined as centres of high biodiversity that are under threat), with loss of traditional knowledge the threat most commonly reported.

**Conclusions** It is concluded that there is a great wealth of ND therapeutic potential in medicinal plants, with promise as drug leads to target multiple hallmarks of ND pathologies. The toolkit methodology appears to be useful in providing a wide reach in the search of this potential.

However, there is an urgent need to preserve the knowledge of ethnomedical use, as well as the habitats on which this knowledge depends.

**KEY WORDS**

Ethnomedicine, medicinal plant, memory, neurotoxicity, neurodegeneration, neuroinflammation, paralysis, rare diseases

**INTRODUCTION**

The various neurodegenerative diseases (NDs) are amongst the most recalcitrant against modern pharmacological interventions, with a failure of treatments to reverse and cure disease progression in Alzheimer's disease (AD) (1), multiple sclerosis (MS), (2), amyotrophic lateral sclerosis (ALS) (3), Huntington's disease (HD) (4) and more rare NDs such as Friedreich's ataxia (5) and Cockayne syndrome (CS) (6). In motor neurone disease, for instance, from the onset of symptoms there is a catastrophic decline to death typically between three to five years (7). Some NDs have a common prevalence worldwide. For instance, the global incidence of AD and other dementias is 43·8 million and rising, and a cause of 2·4 million deaths annually (8). Therapeutic strategies to prevent degeneration for NDs such as Parkinson's disease (PD) and multiple system atrophy (MSA) remain limited due to lack of knowledge of the precise mechanisms underlying the observed pathology (9). Although some NDs are considered to be diseases of aging, there are also paediatric disorders such as Batten disease (10). The search therefore remains for effective curative agents. The general aim of this study is to determine plants with ND therapeutic potential from online-accessible publications of ethnomedical literature. There are three rationales for this. Firstly, of the new therapeutic drugs approved by the U.S. Food and Drug Administration and similar organizations in several of the years from 1981-2019, 50 % of all approvals were natural products or originating from them, including plants (11, 12). Examples of prominent plant-derived drugs include the anti-malarials artemisinin (discovered in *Artemisia annua*) and quinine (from *Cinchona officinalis)* (13) and the anti-cancer agents vincristine and vinblastine (from *Catharanthus roseus*) (14). The anti-leukemic vincristine, for instance, reduces the white blood cell count,

increasing the survival rate of children with leukemia from 20 % to 80 % since the 1950s (15). However, of the 350,699 known plant species according to the Kew Plant List (16), only 6 % have been studied for any biological activity (17, 18) (19). Although this percentage will now be higher due to more recent research, it is likely that the bioactivity of the majority of plant species remain to be investigated (20), and this represents a huge untapped resource. Secondly, of the most useful drugs derived from plants, 80 % were discovered by follow-up of ethnomedical uses (plants used in traditional medical practices) (19). Thus screening indigenous community ethnomedicine data can increase the “hit rate” for discovery of novel active compounds (21). This is because it is the application of a knowledge-based strategy to detect therapeutic potential, in contrast with screening of natural compounds at random, the latter having a low hit rate for identification of relevant bioactivity (20). Thirdly, ethnomedicine has the advantage of demonstrating therapeutic activities of relevance in human populations, with knowledge of efficacy often long-established over many generations. Therefore drugs derived from ethnomedicine are likely to be safer than active compounds with no history of human use (19).

The existing literature often has an emphasis on particular NDs, especially AD. For instance, one study documented 152 plant species with traditional uses for age-related brain diseases, focusing on AD and dementia (22). Some studies focus on a few well-known species, sometimes giving little reference to the underlying pathologies, although other studies (e.g. (23)) do assess the molecular mechanisms which the plants are aimed to target. There are numerous reviews of plants with uses suggestive of alleviation of ND pathology but again the popular focus is on the most common NDs such as AD (e.g. (24)), and PD (e.g. (25)).

Therefore this study has a number of more focused aims. Firstly ND pathologies and symptoms were assessed, from which therapeutic categories could be constructed which could be recognized by ethnomedical practitioners. The rationale for this is to attain a wide set of relevant terms to facilitate the mining of ND therapeutic data. Since the literature appears to report little application of medicinal plants for many of the more rare NDs, this study also included appraisal of pathologies of more rare NDs such as motor neurone disease, Friedreich’s ataxia and Alpers-Huttenlocher syndrome. A further rationale is that although some hallmarks and symptoms, such as memory

impairment, are easily recognised by both clinicians and ethnomedical practitioners, certain hallmarks such as neurotoxicity, of central importance in numerous NDs, cannot be easily translated into terms in ethnomedical use. Anti-neurotoxic effects in plants may indeed exist, which could be revealed from a wider probing of the many medicinal effects reported.

A second aim was to apply these therapeutic categories as a toolkit to find plant uses (identifiable with terms used by ethnomedical practitioners) to alleviate pathologies and symptoms which are also evident in NDs. This tests the hypothesis that the plant kingdom may possess a large reservoir of ND potential. Sources were obtained from online searches, primarily of ethnomedical survey literature.

The findings of this study were then analysed in their ecological contexts. The third aim was to determine factors (such as importance of ethnomedicine to the community) that may affect the richness or weight of ND therapeutic potential. The fourth aim was to assess how this ethnomedical data may be at risk. The world's greatest biodiversity hotspots (BDH) are centres of high biodiversity that are under threat, having lost at least 70 % of their primary native vegetation (26) (27). The survey locations were mapped to establish how many were located in these BDHs and therefore at elevated risk. The surveys were examined systematically to discover what the threats were of most concern to the authors, to inform responses appropriate to how any valuable ethnomedical data of ND potential can be preserved.

**METHODS**

A review of literature was performed using the databases PubMed and Google Scholar, conducted from October 2017 to August 2020, with no time limits on the years of publication. Additional sources of data included reference lists of included articles. The first search enabled an assessment of ND pathologies and symptoms, in order to construct therapeutic categories recognized by ethnomedical practitioners. The search terms of numerous neurodegenerative diseases were applied in relation to terms such as pathology and symptoms (e.g. Alzheimer's disease OR Parkinson's disease OR Multiple Sclerosis AND pathology). A second search consisted of finding ethnomedical surveys containing species with reported ethnological uses

which could also be of ND therapeutic potential. The search terms were: ethnomedical survey, ethnobotanical survey, medicinal plants, medical herb, ethnobotany OR indigenous tribe. These search term alternatives were then also combined with country-specific searches to find further surveys not revealed in the initial searches. . The therapeutic categories were then applied as a toolkit to find plant species with reported therapeutic effects of ND relevance, mined from 157 ethnomedical surveys and 2 ethnomedical databases selected according to the following criteria.

***Inclusion/exclusion criteria***

Survey publications were evaluated for inclusion using the Preferred Reporting Items for Systematic Reviews and Meta-Analyses tool (PRISMA) (28) according to the criteria shown in the flow chart (additional Figure 1). Publications were included which contained data of potential relevance to ND treatment; there was some indication of accurate identification of plant specimens by botanical specialists and/or collection and comparison with species identified in herbaria; and published in the English language.

The following publications were excluded: studies of primarily veterinary significance; surveys with no data of relevance for ND therapeutic potential; studies in which species were selected according to validation by bioactivity reports; studies without evidence of correct identification of species.

Species within publications were excluded from the surveys for any of the following: reports of serious toxicity; no data of relevance for ND therapeutic potential; and vague/ambiguous description of symptoms. Since the aim was to determine plants with ND therapeutic potential, species used in therapies including animal and/or mineral parts were also excluded.

The study was limited to only assessing surveys that primarily reported ethnomedical uses rather than ones integrating the uses with bioactivity studies (ie in which the species were only selected if bioactivity studies validated the ethnomedical report). The results from this study were then used to find the extent to which the ethnomedical reports are confirmed by bioactivity reports. These findings are published separately in a companion paper.

***Data extraction***

Data from 115 ethnomedical surveys not failing the exclusion criteria were mined for pathologies and reported beneficial uses relevant to NDs. A further 42 ethnomedical surveys were mined only for reports of species with memory improvement and anti-paralytic therapeutic use, along with a review of ethnomedical reports of anti-paralytic plants in India. Thus 157 surveys were studied in total. In addition, two ethnomedical databases were accessed for reports of memory improvement and anti-paralytic therapeutic use: the Prelude database of medicinal plants in Sub-Saharan Africa (29) and the Native American Ethnobotany database (30). Data extracted included species names, reports of ethnomedical uses, parts of plants used and location of studies.

***Taxonomy***

Species names, attributions and current family status were checked according to the Kew science online medicinal-plant-names-services (31), the Kew Plant list (16), World Flora Online (32) and Kew Plants of the World Online (33) in order to establish the current accepted scientific name. However, species reported in the literature which are now synonyms according to the above checklists were also included in the results (Additional Table 1) according to their currently accepted species name, with the synonym added. If such synonyms had a number of possible attributions, the species with the highest confidence was selected (as, for instance, with *Aloysia citriodora* Paláu Syn: *Aloysia triphylla* (L'Hér.) Britton). Potential errors were resolved according to the most plausible accepted name. For instance, *Alternanthera brasivine* (L.) Kuntze (as reported by Ribeiro and colleagues (34) was not traceable in the checklists, but the common name (in this case, Terramicina, is suggestive of the currently accepted species *Alternanthera brasiliana* (L.) Kuntze.

***Mapping of survey locations***

The RStudio package (35) was used to map the locations of the studies, using the rgdal, rgeos, dplyr and ggplot2 packages. Biodiversity hotspots spatial data was obtained from the biodiversity hotspots dataset (36). Locations of the studies were determined from maps included in the surveys, and where necessary precise coordinates were obtained using LatLong online geographic tool (37).

## RESULTS AND DISCUSSION

### 1. AN ETHNOMEDICAL TOOLKIT: THERAPEUTIC CATEGORIES

Firstly, from the assessment of ND pathology and clinical symptoms, a list of desirable therapeutic categories was composed which can be translated into terms used in ethnomedicine where necessary, although some clinical categories and ethnomedical terms are equivalent (Table 1). The categories were restricted to those that could be recognized by ethnomedical practitioners, so therapeutic effects such as anti-oxidants and anti-amyloid aggregation were not included.

**Table** 1 An ethnomedical toolkit: ND pathology/symptom hallmarks mapped to therapeutic categories used in ethnomedicine

| ND Disease category | Hallmarks of pathology and symptoms | Therapeutic category [Ethnomedical category of therapeutic potential] |
|---|---|---|
| Neurotoxicity | Misfolding + aggregation of proteins: e.g. amyloid β (Aβ) and tau in AD; alpha-synuclein (α-syn) in PD and MSA; superoxide dismutase 1 (SOD1) + TAR DNA binding protein (TDP-43) in ALS; Huntingtin in HD; prion proteins in Spongiform encephalopathies (38). These cause neurotoxic effects by disrupting synaptic transmission, damaging mitochondria and impeding proteasomal clearance (39). Malfunctioning microglia and astrocytes (neuron support cells) release neurotoxic factors that kill neurons (40). | Detoxifying/poison antidote, anti-venom |
| Memory and cognitive impairment | Memory and cognitive impairment occurs in AD, A-H, BD, CS, FRDA, , HD, MSA, MS, NPC, PD, VD, but also non-ND diseases + traumatic causes (41). May result from neurotoxicity [see neurotoxicity box] and its effects, such as disrupted synaptic transmission + neuronal death. | Memory/cognitive improvement, anti-dementia |
| Paralysis | Degeneration of motor neurons results in paralysis: in ALS neurotox-icity [see neurotoxicity box] leads to axonal degeneration + myelin loss in spinal cord (7); in MS is associated with neurodegeneration of neuronal myelin sheath (42); in FRDA a mutation results in CNS effects including paralysis (43); in polio virus infection can lead to spinal motor neuron loss + paralysis, + in postpolio syndrome there is further degeneration of neuromuscular units (44). | Anti-paralytic/hemiplegia/ paraplegia/polio |
| Parkinson's disease/Parkinsonism | Associated with misfolded α-synuclein aggregation, resulting in dopaminergic neuron loss, leading to motor symptoms e.g. rigidity, resting tremor and postural instability(45). | Anti-PD, anti-tremor |
| Neuroinflammation | Neuroinflammation may drive abnormal aggregation of misfolded proteins (46). A persistent inflammatory response, possibly induced by dysregulated cytokine proteins, perturbs microglia and astrocytes to become neurotoxic (47). | Anti-inflammatory |
| Impaired neurogenesis | Neurogenesis (the production of new neurons) does occur in the adult brain but is impaired in NDs and may contribute to the disease process (48). | Wound healing, anti-ulcer |
| Immune involvement | Triggers of immune dysfunction can contribute to synaptic and neuron loss and drive ND progression (49). Dysfunctional immune system: in MS attacks myelin sheath of CNS neurons; and is implicated in AD, HD, glaucoma + BD. | Boost immune system, immunomodulation |
| Mitochondrial dysfunction | Mitochondrial dysfunction or damage results in energy failure and fatigue. This is a pathological hallmark of NDs including AD and HD (50), ALS (7), MSA (51), BD (52), Gangliosidosis (53), peripheral neuropathies (54), FRDA (43) + A-H (55). | Anti-fatigue/weakness, /boosting energy/strength |
| Infectious agents | Microbial agents have been implicated in inducing hallmarks of ND such as misfolded protein aggregates, in AD and possibly ALS. | Anti-microbial |

| | | |
|---|---|---|
| | Evidence includes pathogens colocalizing with AD pathology and anti-virals blocking Aβ and tau pathology (56). | |
| Aging | Risk factors associated with NDs include hallmarks of aging: e.g. genomic instability, telomere attrition, epigenetic alterations, impaired proteostasis, mitochondrial dysfunction, cellular senescence and stem cell exhaustion (57). | Anti-aging, promoting longevity |
| Vascular disease and hypertension | The majority of AD patients display vascular involvement. Atherosclerotic vascular wall thickening impedes $O_2$ and nutrient delivery to the brain, leading to neuronal loss (58). Hypertension raises brain amyloid and tau deposition (59) and impairs cerebral blood vessels, leading to brain ischemic damage and impaired cognitive function (60). | Anti-hypertensive, cardiotonic, anti-thrombotic, anti-atherosclerotic, anti- stroke |
| Lysosomal storage diseases e.g. Gaucher + Batten disease | Enzyme deficiencies in lysosomes (which degrade cell waste) lead to toxic accumulation of macromolecules e.g. of unesterified cholesterol in NPC (61). Neurons are vulnerable to accumulating waste, leading to neurodegeneration (62). | Anti-hyperlipidemia; cholesterol-lowering |
| Non-ND neuro-logical conditions | Agents alleviating these conditions can pass through the blood-brain barrier and have a therapeutic effect (63) before being metabolized: thus may be of translational relevance in NDs. | Anti-epileptic, anxiolytic, alleviation of other neurological conditions |

Abbreviations: AD, Alzheimer's disease; A-H, Alpers-Huttenlocher syndrome; ALS, Amyotrophic lateral sclerosis; ALS BD, Batten disease; CS, Cockayne syndrome; FRDA, Friedreich's ataxia; HD, Huntington's disease; MSA, Multiple system atrophy; MS, Multiple sclerosis; NPC, Neimann-Pick disease type C;PD, Parkinson's disease; VD, Vascular dementia.

These toolkit categories were then applied to find plant therapeutic effects of potential ND relevance reported in ethnomedical terms, indicated as follows.

## 2. APPLICATION OF THE TOOLKIT TO FIND PLANTS WITH ND THERAPEUTIC POTENTIAL

A total of 157 ethnomedical surveys that passed the inclusion criteria were obtained from the various online-accessible literature searches. Two databases were also mined for species with ND therapeutic potential. The surveys represented 67 countries spanning all of the inhabited continents, to obtain as global a reach as possible of therapeutically relevant data.

The 157 surveys and databases were then mined to determine which plant species demonstrated any beneficial effects in these therapeutic categories. 2001 plant species were found to have a reported ethnomedical use of ND therapeutic potential (summarized in Table 2).

**Table 2** Summary of number of species with reported therapeutic effects, and relevance to various NDs

| Therapeutic category | Number of species | ND-specific relevance |
|---|---|---|
| Detoxifying/poison antidote/anti-venom | 124 | AD, ALD, ALS, CJD, FTD, GLAU, HD, MSA, MSP, PD |
| Memory/cognitive improvement/dementia | 130 | AD, A-H, BD, CS, FRDA, HD, MSA, MS, NPC, PD, VD |
| Anti-paralytic/ hemiplegia/ paraplegia/ polio | 183 | ALS, MS |
| Anti-PD/ anti-tremor | 14 | PD, PS |
| Anti-inflammatory | 974 | Most/all NDs |
| Wound healing/anti-ulcer | 110 | Most/all NDs |
| Boost immune system/immunomodulation | 34 | MS, AD, HD, GLAU, BD |
| Anti-fatigue/weakness/boosting energy/strength | 49 | AD, ALS, A-H, ALS, HD, GLAU |

| | | |
|---|---|---|
| Total anti-microbial all types [anti-bacterial, anti-viral, anti-fungal] | 1176 | AD, ALS |
| Anti-aging/promoting longevity | 20 | AD, ALS, PD, VD |
| Anti-hypertensive, cardiotonic, anti-thrombotic, anti-atherosclerotic, anti-stroke/traumatic brain injury | 246 | AD, VD |
| Anti-hyperlipidemic/anti-hypercholesterolaemic | 35 | AD, NPC |
| Anti-epileptic/anxiolytic/alleviation of other neurological conditions | 77 | Most/all NDs |

Abbreviations: *AD, Alzheimer's disease; ALD, Alexander disease; A-H, Alpers-Huttenlocher syndrome; ALS, Amyotrophic lateral sclerosis CJD, Creutzfeldt-Jakob disease; FTD, Frontotemporal dementia; GLAU, Glaucoma; HD, Huntington's disease; MSA, Multiple system atrophy; MS, Multiple sclerosis; MSP, Multisystem proteinopathy; ND, neurodegenerative disease; NPC, Neimann-Pick disease type C; PD, Parkinson's disease; PS, Perry syndrome; SMA, Spinal muscular atrophy; VD, Vascular dementia.*

A detailed list of ethnomedical uses for each of these species, along with the updated taxonomic attribution and status, locations of ethnological surveys, parts of plant used and references can be found in the Additional Table 1.

Ethnomedical uses were reported for every ND therapeutic category searched for (Table 2). This gives the prospect that these species could be targeted to several of the hallmarks of pathology implicated in NDs, and that there is a rich pool of species with such potential. These findings are detailed as follows, for each therapeutic category.

### *2.1 Detoxifying/poison antidote/anti-venom (anti-neurotoxicity)*

Normal biological function of proteins depends upon their folding into the correct three dimensional structure. A key hallmark of pathology in numerous NDs is the misfolding and aggregation of specific proteins, which are toxic to neurons (64). For instance, amyloid proteins are toxic by disrupting synaptic transmission, damaging mitochondria and impeding proteasomal clearance (the cell machinery for removing neurotoxic proteins) (39). There are other sources of neurotoxicity cited for NDs: for instance, microglia and astrocytes (which normally provide support to neurons) instead malfunction, releasing neurotoxic factors (40) by mechanisms involving nitric oxide, glutamate, or unknown neurotoxins (65) (66) (67). Thus of potential relevance to these pathologies are the ethnomedical reports of plants with detoxifying/poison antidote/anti-venom beneficial effects, comprising 124 species (Table 2, Supplementary Table 1). These plants could have the capacity to neutralize the neurotoxic factors, driving numerous ND pathologies such AD, ALS, HD, PD AD, ALD, ALS, CJD, FTD, glaucoma, HD, MSA, MSP, PD and LSDs.

### *2.1 Memory and cognitive impairment*

Amnestic syndromes are common in AD, vascular dementia, PD, HD, epilepsy, MS, ALS, and paediatric NDs such as Batten disease; traumatic and toxic causes (41). The primary drivers of neurodegeneration in AD appear to be toxic soluble amyloid β oligomers cooperating with pathological tau protein, along with mitochondrial damage, to progressively degenerate the learning and memory circuitry (68). Thus the 130 plant species with ethnomedical reports of memory/cognitive improvement (summarized in Table 3) could be of therapeutic relevance to target the pathologies underlying memory dysfunctions associated with NDs.

**Table 3** Plant species with examples of important ethnological uses of ND therapeutic potential

| **Therapeutic effect** | **Species** |
|---|---|
| Memory/cognitive improvement /dementia | *Acanthospermum hispidum, Acer monspessulanum, Acokanthera schimperi, Adansonia digitata, Aframomum melegueta, Albizia zygia, Allium fistulosum, Allium sativum, Alstonia boonei, Angraecum eichlerianum, Annona muricata, Annona senegalensis, Asystasia gangetica, Bacopa floribunda, Bacopa monnieri, Bambusa vulgaris, Baphia nitida, Bobgunnia madagascariensis, Borago officinalis, Boswellia sacra, Bridelia ferruginea, Brillantaisia owariensis, Bunium persicum, Capparis erythrocarpus, Carica papaya, Carissa edulis, Centella asiatica, Cinnamomum verum, Cissampelos owariensis, Cissus aralioides, Citrus aurantiurn, Cleome gynandra, Cocos nucifera, Codonopsis clematidae, Cola acuminata, Combretum micranthum, Crossopteryx febrifuga, Curculigo pilosa, Cuscuta americana, Cymbopogon citratus, Cymbopogon densiflorus, Cymbopogon giganteus, Cynanchum viminale, Cynodon dactylon, Dalbergia lactea, Detarium microcarpum, Digitaria debilis, Dioscorea mangenotiana, Dipteryx alata, Dysphania ambrosioides, Ehretia cymosa, Elaeis guineensis, Eleutherine bulbosa, Emilia abyssinicus, Entandrophragma utile, Erythrina abyssinica, Erythrina senegalensis, Euphorbia hirta, Evolvulus alsinoides, Ficus carica, Ficus exasperata, Ficus platyphylla, Flueggea virosa, Fragaria nubicola, Galinsoga parviflora, Harungana madagascariensis, Heliotropium indicum, Heteropterys tomentosa, Hippophae rhamnoides, Hymenocardia acida, Ipomoea batatas, Jatropha curcas, Lannea acida, Lawsonia inermis, Leonurus cardiaca, Lippia multiflora, Litsea cubeba, Maesa lanceolata, Mangifera indica, Melissa officinalis, Mentha arvensis, Mitragyna inermis, Momordica balsamina, Momordica charantia , Mondia whitei, Morinda lucida, Moringa oleifera, Musa paradisiaca, Nauclea latifolia, Ocimum americanum, Ocimum tenuiflorum, Papaver dubium, Papaver rhoeas, Parinari curatellifolia, Parinari excelsa, Pergularia daemia, Phyllanthus amarus, Picralima nitida, Piper guineense, Pistacia atlantica, Plumbago zeylanica, Prunus africana, Pseudocedrela kotschyi, Psychotria elata, Punica granatum, Quassia undulata, Rhodiola tibetica, Rhytidocaulon macrolobum, Rosmarinus officinalis, Saccharum officinarum, Schwenckia americana, Scolymus hispanicus, Scoparia dulcis, Securidaca longepedonculata, Solanum incanum, Tamarindus indica, Tephrosia purpurea, Tetrapleura tetraptera, Trifolium repens, Uraria picta, Vernonia colorata, Vitellaria paradoxa, Vitex madiensis, Vitex negundo, Vitex simplicifolia, Vitis vinifera, Zanthoxylum zanthoxyloides, Zea mays, Ziziphus mucronata, Zingiber officinale* |
| Anti-paralytic/ hemiplegia/ paraplegia/ polio | *Abutilon indicum,* Acacia ehrenbergiana, *Acacia etbaïca, Acacia modesta, Achyranthes aspera, Acokanthera schimperi, Acridocarpus orientalis, Adansonia digitata*, *Adenostoma sparsifolium, Aframomum melegueta, Albizia lebbeck, Allium sativum, Alstonia macrophylla, Alstonia scholaris, Amaranthus viridis, Anacyclus pyrethrum, Annona senegalensis, Anthocleista djalonensis, Aquilaria malaccensis, Ardisia gigantifolia, Aristolochia grandiflora, Artemisia absinthium, Asparagus africanus*, *Asparagus racemosus, Asphodelus aestivus, Azadirachta indica, Barnadesia arborea, Bergenia ciliata, Blumea balsamifera, Boerhavia diffusa, Boesenbergia rotunda, Bombax ceiba, Breynia vitis-idaea, Bridelia ferruginea, Brugmansia x candida, Buddleja mendozensis, Cajanus cajan*, *Calotropis procera*, *Capparis grandis, Capparis spinosa, Caralluma tuberculata, Cardiospermum halicacabum, Cassia fistula*, *Castilleja coccinea, Cayratia japonica, Celastrus paniculatus, Celosia argentea, Celtis toka, Cissus adnata, Citrus aurantifolia, Citrus maxima, Clerodendrum paniculatum, Clinopodium umbrosum, Cola acuminata, Colubrina asiatica, Cornus sericea, Crassocephalum vitellinum, Crinum nubicum*, *Croton mubango, Croton tiglium, Cucurbita pepo, Cuminum cyminum, Cyanthillium patulum, Cymbopogon citratus, Cymbopogon giganteus, Cymbopogon nardus, Cymbopogon proximus, Cyperus rotundus, Daniellia oliveri, Dioscorea alata, Dracaena spicata*, *Ehretia cymosa, Embelia ribes, Entada pursaetha, Eryngium caeruleum, Eryngium creticum, Erythrina senegalensis, Euclea racemosa, Euphorbia abyssinica*, *Euphorbia kamerunica, Euphorbia pallens, Euphorbia terracina, Ficus thonningii, Glyphaea brevis, Gossypium barbadense,* Guilandina bonduc, *Harpullia sp., Heliconia psittacorum, Helinus integrifolius, Heliotropium indicum, Hernandia ovigera, Hibiscus fuscus, Holoptelea integrifolia, Hoya parasitica, Impatiens stuhlmannii*, *Ipomoea pes-caprae*, *Iresine diffusa, Jasminum grandiflorum, Jasminum syringifolium, Jatropha curcas*, *Jatropha glandulifera, Jatropha gossypifolia, Juniperus recurva, Justicia* |

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|---|---|
| | *gendarussa, Kalanchoe pinnata,* Kleinia longiflora, *Laurelia sempervivens, Lens culinaris, Litsea cubeba, Lomatium californicum, Lonchocarpus cyanescens, Lonchocarpus laxiflorus, Lonicera involucrata, Melilotus suaveolens, Millettia eetveldeana, Mitragyna rubrostipulata, Moneses uniflora*, *Morinda citrifolia, Moringa oleifera, Mucuna pruriens, Nauclea latifolia*, *Neopicrorhiza scrophulariiflora, Ocimum tenuiflorum, Opuntia dillenii*, *Origanum vulgare, Paederia foetida, Paullinia pinnata, Pentadiplandra brazzeana, Persea americana, Phlogacanthus thyrsiformis, Phragmanthera usuiensis, Phyllanthus amarus, Physalis minima, Pimpinella anisum, Pinus contorta, Plumbago indica, Plumbago zeylanica, Pongamia pinnata, Portulaca oleracea, Primula vulgaris, Protium glabrescens, Prunus amygdalus, Prunus cerasoides, Pseudotsuga menziesii, Pupalia lappacea, Raphanus raphanistrum, Ribes sp.*, *Rosa sp.,* Rotheca myricoides, *Rourea coccinea, Rubus sp., Ruta chalepensis, Ruta montana, Saccharum officinarum, Salix denticulata, Salvia tubiflora, Satureja thymbra*, *Scaevola taccada, Semecarpus anacardium*, Senna alata, *Senna italica, Sida acuta, Sida cordata, Sigesbeckia orientalis,* Solanecio mannii, *Solanum aethiopicum, Solanum dasyphyllum, Sorghum bicolor*, *Spilanthes acmella, Stereospermum kunthianum, Symphyotrichum puniceum*, *Tephrosia purpurea*, *Thonningia sanguinea, Thymbra capitata, Thymbra spicata, Tinospora caffra, Trema orientalis, Urtica dioica, Withania somnifera, Zanthoxylum capense, Zea mays*, *Zingiber montanum, Zingiber officinale* |
| Anti-PD/anti-tremor | *Acanthospermum hispidum, Allium cepa, Brillantaisia owariensis, Calotropis procera*, *Cecropia pachystachya, Chiliadenus iphionoides, Citrus aurantifolia, Gardenia ternifolia, Lens culinaris, Lippia multiflora, Minthostachys glabrescens, Mucuna pruriens, Ottelia ulvifolia, Securidaca longipedunculata* |
| Anti-hyperlipid-emic, cholesterol-reducing | *Allium cepa, Allium rubellum, Aphelandra pilosa, Artocarpus altilis, Aspidosperma subincanum*, *Berberis integerrima, Bixa orellana*, *Borago officinalis*., *Buchenavia tomentosa, Camellia sinensis, Cichorium intybus, Citrullus colocynthis, Crataegus spp., Cucumis anguria, Cynara scolymus*, *Cyphomandra betacea, Dipteryx alata* , *Eriobotrya japonica*, *Jatropha elliptica, Juglans neotropica, Lavandula stoechas, Morus nigr*a, *Olea europaea, Opuntia ficus-indica, Passiflora quadrangularis, Prunus dulcis, Psittacanthus calyculatus, Rosmarinus officinalis, Simaba ferruginea*, *Verbena litoralis, Vochysia rufa* |

### *2.3 Anti-paralytic/ hemiplegia/ paraplegia/ polio*

Paralysis is a chief symptom in NDs such as ALS, spinal muscular atrophy (SMA) and Friedreich's ataxia. The causes of paralysis are specific to the particular ND. In ALS, neuronal aggregation of misfolded proteins, neuroinflammation, astrocyte and microglial activation, mitochondrial dysfunction and other perturbed mechanisms result in axonal degeneration and loss of myelin in the lateral spinal cord, leading to paralysis (7) (69). In MS, either an inflammatory attack of CNS targets or degeneration within the myelin sheath of neurones lead to axonal loss and the resulting paralysis (42). In SMA the paralysis results from loss of the survival motor neuron-1 (*SMN1*) gene. In Friedreich's ataxia, mutations in the *FXN* gene (a mitochondrial protein) lead to CNS effects such as dorsal root ganglia neuron destruction, resulting in paralysis (43). Of relevance to these NDs, in this study 183 species were found with documented anti-paralytic ethnological use (summarized in Table 3) which could alleviate the motor paralysis central to ALS, MS and SMA, although these could also have been reporting therapeutic affects for other conditions such as spinal cord injury.

### *2.4 Anti-PD/ anti-tremor*

PD is associated with misfolded α-synuclein aggregation (70), resulting in dopaminergic neuron loss (45). This leads to motor symptoms such as rigidity, resting tremor and postural instability, along with non-motor symptoms such as cognitive deficits. There are several other tremor syndromes, each determined by the dysfunction of specific neural circuits. However, tremor is the cardinal symptom of PD, and asymmetric rest tremors seen in the leg are almost always due to PD (71). Of potential therapeutic relevance are the 14 plant species with reports of anti-tremor ethnological use (Table 3).

### *2.5 Anti-inflammatory effects*

Although various NDs share a trait of abnormal aggregation of misfolded proteins (72), there is evidence that neuroinflammation may drive these effects (reviewed by (46). Inflammation is a host response to insults such as injury or infectious agents, resulting in the influx of cells from the immune system, and induction of inflammatory mediators such as cytokines into the affected area. Cytokines are small proteins secreted by cells such as microglia. They trigger signalling cascades leading to upregulation and/or downregulation of genes and production of further cytokines. So-called proinflammatory cytokines are ones that promote inflammation (73) (74) and others are involved in anti-inflammatory pathways (75). Dysregulation of the cytokines may drive pathologies via perturbing the microglia phenotype (76).

Microglia are non-neuronal immune cells within the CNS. They exhibit a sentinel function for surveillance of their environment; a housekeeping function to maintain normal neuronal operation and a warrior state function, responding to threats such as infectious pathogens and injurious-self proteins (77). Dysregulation of these functions results in an imbalance that may initiate or propagate neurodegeneration (47). Astrocytes are another non-neuronal cell with activities such as shuttling of neurotransmitter and cerebrovascular regulation (78). In NDs, activated pro-inflammatory microglia induce astrocytes to be reactive, losing the ability to promote neuronal survival and instead induce neuron death. When the formation of reactive astrocytes is blocked, neuron death is prevented (79).

In the healthy brain, initial inflammatory responses are self-limited once the stimulus is terminated. If, however, the insult persists for a long period this may result in permanent activation of microglia and their constant release of proinflammatory cytokines, with neurotoxic consequences. Chronic neurodegenerative diseases, such as AD, MS, PD, ALS, peripheral neuropathies (e.g. Charcot–Marie–Tooth disease) and glaucoma are all associated with chronic neuroinflammation (80), in which the microglial activation is persistent. Neuroinflammation contributes to the pathogenesis and/or disease progression in paediatric NDs such as (Batten disease) (81), Gangliosidosis (53) and Niemann-Pick disease (61). Thus correcting the microglial dysregulation may be a potential therapeutic target (47). For instance, microglial silencing reduces clinical severity of MS, suggesting their involvement in CNS damage processes (82). Anti-neuroinflammatory therapies using non-steroidal anti-inflammatory drugs (NSAIDs) have been disappointing (76). However, there is a marked reduction in the incidence of AD in individuals with rheumatoid arthritis (83), which is probably the result of long-term anti-inflammatory therapy. There is also an inverse association between NSAID use and AD (84), with NSAIDs reducing the risk of developing AD or PD by as much as 50 % (85). This evidence suggests that targeting neuro-inflammation is a viable therapeutic strategy.

Thus the 974 plant species with reported anti-inflammatory ethnological use could be of therapeutic relevance to these various inflammatory aspects of ND pathologies (Table 2, Additional Table 1).

### *2.6 Wound healing/anti-ulcer (for neurogenic potential)*

It was previously believed that the adult brain is incapable of producing new neurons (reviewed by (86). However, neurogenesis (the process by which new neurons are formed) does persist in the adult brain in localized neurogenic niche regions (87) including in humans (88) but is impaired in NDs and may even contribute to the disease process (48). Inflammation may play a role in this, by inhibiting neurogenesis in these neurogenic niches (89). Therapeutic approaches in animal models aim at potentiating neuronal regeneration, by suppressing growth inhibitory signals, providing growth promoters, and enhancing neuron growth programs (90). Thus the enhancement of neurotrophic factors may yield therapeutic benefits in NDs. Therefore plants with ethnomedical

reports of wound healing/ anti-ulcer properties (110 species) could be a source of neurotrophic or neurogenic factors to harness in therapeutic strategies aimed at potentiating neural regeneration (Table 2, Additional Table 1).

### *2.7 Boost immune system/immunomodulation*

It is now recognised that a perturbed immune response forms an integral part of neurodegeneration, being a central player in disease onset and driving its progression (reviewed by (85) (49). Research efforts are focused on identifying triggers of immune dysfunction which can contribute to synaptic and neuron loss and build-up of pathogenic proteins. The immune system appears to particularly underpin MS, in which auto-reactive T cells, macrophages and B cells cause an inflammatory attack of CNS targets, notably myelin (91). The immune system is also implicated in AD, HD, glaucoma, and Batten disease. Of potential relevance to these pathologies are the 34 plant species with documented immune system boosting or immunomodulation ethnological use (Table 2, Additional Table 1).

### *2.8 Anti-fatigue/weakness/ energy-boosting*

Mitochondrial dysfunction or damage, resulting in energy failure, is a pathological hallmark of a number of NDs, including AD and HD (50), ALS (7)), MSA (51), Batten disease (52), Gangliosidosis (53), peripheral neuropathies (54), Friedreich's ataxia (43) and Alpers-Huttenlocher syndrome (55). Moreover, anatomical destruction of brain circuits can occur in NDs. For instance, in PD, basal ganglia-thalamocortical circuit dysfunction is the anatomical correlate of fatigue symptoms, in which patients fatigue rapidly in motor tasks (92).

Inflammation may be an additional driving force for fatigue (93). In patients undergoing cancer treatment (Liu et al., 2012) and in CFS/ME patients inflammatory markers such as pro-inflammatory Il-6 levels are associated with fatigue symptoms (94).

Of therapeutic potential to these ND-related fatigue symptoms are the 49 plant species found with reported anti-fatigue/energy-boosting ethnological uses (Table 2, Additional file 1).

### *2.9 Anti-microbial effects*

Molecular hallmarks of neurodegeneration (such as misfolded protein aggregates, synaptic impairment, and neuronal death) may be induced by microbial agents, amplified by risk factors such as aging (95). Itzhaki and colleagues (56) have also summarized evidence in favour of an infectious agent in AD (e.g. pathogen signatures specifically colocalize with AD pathology) and may have a causative role in the pathology. For instance, antivirals such as acyclovir block HSV1-induced Aβ and tau pathology *in vitro* (96). Microbes implicated in AD include *Herpes simplex* virus type 1 (HSV1) and type 2 (HSV2), *Chlamydia pneumoniae*, and several spirochaete and fungal species (56). Microbes may also act on pathways implicated in motor neuron degeneration (97). Of significance to these infectious agents implicated in NDs are the 1176 plant species found with documented anti-microbial ethnological use (Table 2, Additional file 1).

### *2.10 Anti-aging/promoting longevity*

Age is the main common risk factor in NDs such as AD and PD, although the reasons are not clear (98). A number of hallmarks of aging have been identified, such as genomic instability, telomere attrition, epigenetic alterations, impaired proteostasis, mitochondrial dysfunction, cellular senescence and stem cell exhaustion (57). Therapies targeting such mechanisms are being investigated, aiming to extend the healthy years of life, with the social and economic benefits it would create in a more active and vibrant older population (99).

Changes in these mechanisms are common to a number of NDs. For instance, axonal degeneration occurs at early stages of AD, ALS and PD before the onset of clinical symptoms (100). This may be due to increasing mitochondrial dysfunction, or toxic proteins and DNA damage accumulating with time (reviewed by (98).

Thus the 20 plant species with reported anti-aging/ longevity-promoting ethnological uses (Table 2, Additional file 1) may be of significance to target these age-related degenerative mechanisms.

### *2.11 Anti-hypertensive, cardiotonic, anti-thrombotic, anti-atherosclerotic, anti-stroke/traumatic brain injury*

In vascular dementia or more precisely vascular cognitive impairment, the dementia is attributed to cerebrovascular as opposed to neurodegenerative pathologies (101). However, both vascular and

neurodegenerative pathologies commonly co-exist, with the majority of AD patients displaying vascular involvement, in which brain amyloid is associated with the cerebral arterial vessels (102). There is atherosclerotic vascular wall thickening and blood vessel occlusion (103), leading to impaired oxygen and nutrient delivery to the brain, which in turn may lead to neuronal loss (58).

Elevated blood pressure and cerebrovascular lesions are associated with increased dementia risk (104) (105); reviewed by (106). Moreover, raised blood pressure is associated with higher brain amyloid burden and greater tau deposition (107) (59). Hypertension impairs cerebral blood vessels, leading to ischemic damage of white matter regions crucial for cognitive function (60). In relation to these pathologies is the therapeutic potential of the 246 plant species with reported ethnological vascular uses, including anti-hypertensive, cardiotonic, anti-thrombotic, anti-atherosclerotic, and anti-stroke/traumatic brain injury effects (Table 2, Additional file 1).

### *2.12 Anti-hyperlipidemic/anti-hypercholesterolaemic effects*

Lysosomes, crucial to degrading cell waste and debris, exhibit enzyme deficiencies in the lysosomal storage diseases (LSDs). This leads to the toxic accumulation of macromolecules that fail to be degraded, such as the lysosomal accumulation of unesterified cholesterol in Niemann-Pick disease (reviewed by (61).

Gaucher disease is an LSD caused by a mutation in the lysosomal β-glucocerebrosidase gene leading to glucosylceramide accumulation (reviewed by (108). Batten disease is a group of devastating paediatric NDs resulting in blindness, cognitive and motor decline, and premature death (109). The pathological hallmarks include intralysosomal accumulation of a protein-lipid mixture termed ceroid (81) (110). Progressive neurodegeneration is a common hallmark of most LSDs, with neurons being particularly vulnerable by being unable to use cell division to reduce the accumulating waste (62). Therefore of therapeutic potential to improve clearance of the accumulating lipids are the 35 plant species with ethnological reports of anti-hyperlipidemic/anti-hypercholesterolaemic effects (Table 2, Additional file 1).

### *2.13 Anti-epileptic/anxiolytic/alleviation of other neurological conditions*

Whilst epilepsy, anxiety, depression and mood disorders are not NDs, medical plants that are reported to alleviate these conditions could, in order for them to be effective within the brain, be demonstrating the capability of their bioactive compounds to pass passing through the blood-brain barrier (BBB) and also for such compounds to have a therapeutic effect before they are metabolized. Not all bioactive compounds contained within a particular plant species of ND therapeutic relevance may pass the BBB. However, there is still the possibility that certain plants may have bioactive compounds with both non-ND and ND-therapeutic effects, suggesting the relevance of including plants with reported therapeutic roles in non-ND neurological conditions. Thus of potential relevance are the 77 plant species with ethnological reported uses of anti-epileptic/anxiolytic effects and alleviation of other neurological conditions.

## 3. APPLICATION OF ETHNOMEDICAL TOOLKIT TO FIND PLANT THERAPEUTIC POTENTIAL TO SPECIFIC NDS

The therapeutic categories were also applied as a toolkit to find ethnomedical uses of plants with therapeutic potential to specific NDs (Table 4). For instance, Alpers-Huttenlocher syndrome is characterized by severe mitochondrial DNA depletion, leading to neuronal loss and severe epileptic seizures (55). Therefore plant species with therapeutic potential include energy boosting, anti-epileptic and wound healing (for neurogenic potential) uses.

**Table 4** Application of ethnomedical toolkit to find ND-specific plant therapeutic potential

| Neurodegenerative disease | Hallmarks of pathology and symptoms | Plant species with therapeutic potential |
|---|---|---|
| Alpers-Huttenlocher syndrome | Severe mitochondrial DNA depletion, leading to neuronal loss and severe epileptic seizures; often resistant to antiepileptic medication (reviewed by (55). | Energy boosting, anti-epileptic; wound healing (for neurogenic potential) |
| Alzheimer's disease | Misfolded amyloid-β and hyper-phosphorylated tau proteins, defective acetylcholine neurotransmitter function + environment-al stressors interplaying with genetic risk factors lead to memory + cognitive deterioration, mitochondrial dysfunction + neuronal death (64). Inflammatory mediators contribute to disease progression (40). Motor dysfunction, ambulation loss, swallowing difficulties + neuropsychiatric changes in late stages (111). | Memory improvement, anti-inflammatory, anti-toxin and energy boosting |
| Charcot-Marie-Tooth disease | The most common peripheral neuropathy. Inherited or acquired factors (e.g. toxin insults and inflammation) lead to peripheral nerve function loss**.** Hallmarks and symptoms include protein aggregation abnormalities, mitochondrial dysfunction, muscle wasting and weakness (reviewed by(54). | Anti-fatigue, wound healing (for neurogenic potential), anti-toxin, anti-inflammatory |
| Cockayne syndrome | An autosomal recessive disorder, marked by severe neurological manifestations: microcephaly and cognitive deficits, neuronal loss and CNS demyelination (6). | Memory/cognitive improvement, wound healing anti-toxin |

| | | |
|---|---|---|
| Friedreich's ataxia | *FXN* gene mutations result in CNS effects including paralysis (reviewed by (43). | Anti-paralytic, anti-toxin |
| Glaucoma | A multifactorial ND of the optic nerve and retinal ganglion cells, leading to blindness (reviewed by (112). Associated with elevated intraocular pressure, with evidence of amyloid β, synuclein, and tau protein deposition, atherosclerosis, neuro-inflammation and immune involvement (reviewed by (113). | Anti-inflammatory, anti-hypertensive, wound healing, anti-toxin and immune modulatory uses |
| Huntington's disease | *HTT* gene mutation leads to neuronal aggregation of misfolded Huntingtin protein (htt), causing neuronal damage in neostriatum and cerebral cortex, later spreading throughout brain (114); oxidative stress; and neuroinflammatory activation of microglia (115). Other hallmarks include mitochondrial dysfunction, reduced energy metabolism and reduced neurogenesis. May include microbial involvement (116). Symptoms include movement chorea (involuntary, jerk-like muscle contractions) cognitive impairment and psychiatric symptoms (117). | Memory/cognitive improvement, anti-paralytic, anti-inflammatory, anti-toxin, energy boosting and wound healing (for neurogenic potential), mood disorder/anti-depressant, anti-microbial |
| Lysosomal storage diseases | Deficiencies in waste-degrading lysosomes lead to accumulation of macromolecules (e.g. of cholesterol in Niemann-Pick disease) which are toxic to neurons (reviewed by (61). | Cholesterol-lowering, anti-hyperlipidemic, anti-toxin |
| Motor neurone disease (MND) | *Amyotrophic lateral sclerosis (ALS):* neuroinflammation, axonal degeneration, loss of myelin in the lateral spinal cord and neuronal aggregation of misfolded proteins, with neurotoxic effects (3) (118). It leads to atrophy, weakness, and muscle paralysis (7).<br>*Spinal muscular atrophy (SMA*): selective degeneration of lower motor neurons due to *SMN1* gene mutations results in progressive muscle weakness and paralysis - the leading genetic cause of infant death (119). | Anti-paralytic, anti-inflammatory, anti-toxin, energy boosting and wound healing (for neurogenic potential) |
| Multiple sclerosis | An autoimmune destructive attack on myelin and neuronal axons, or alternatively may be a primary degenerative disease accompanied by inflammation (91). Commonly early relapsing and remitting episodes, followed by a chronic progressive phase. | Immune modulation, anti-inflammatory, anti-toxin + wound healing (for neurogenesis), anti-paralytic |
| Parkinson's disease | Misfolded α-synuclein aggregation results in dopaminergic neuron loss, associated with inflammation, leading to motor symptoms (e.g. resting tremor) and cognitive deficits (45). | Anti-tremor, anti-inflammatory, memory/ cognitive improvement uses |
| Prion diseases | Transmissible, progressive, fatal brain diseases (e.g. Creutzfeldt -Jakob disease). Abnormally folded prion protein leads to neuroinflammation, microglial and astroglial activation, neuronal damage and loss (reviewed by (120). | Anti-toxin, anti-inflammatory, wound healing (for neurogenic potential) |
| Vascular disease | In AD patients displaying vascular involvement, brain amyloid distribution is associated with cerebral arteries which are also atherosclerotic, leading to impaired brain $O_2$ levels and neuronal loss (58). Hypertension is associated with increased amyloid and tau deposition (107) (59). | Anti-hypertensive, anti-atherosclerotic, cholesterol-lowering, anti-hyperlipidemic |

Another example is HD, in which misfolded Huntingtin protein causes neuronal damage, neuroinflammation, mitochondrial dysfunction, reduced energy metabolism and reduced neurogenesis (115). There may also be a microbial involvement (116). Thus plant species with therapeutic potential include memory/cognitive improvement, anti-paralytic, anti-inflammatory, anti-toxin, energy boosting, wound healing (for neurogenic potential) and mood disorder/anti-depressant uses.

## 4. VALIDATION OF ETHNOLOGICAL REPORTS

Bioactivity reports validating these ethnological reports are too many to report here and thus are the focus of a companion paper (submitted), in which many of the species with reported

ethnological uses have a confirmed bioactivity of ND therapeutic potential. For instance, for *Centella asiatica* the ethnological reports of memory improvement (121) (122) are validated by bioactivity reports of attenuated cognitive deficits (123), anti-amyloidogenic (124) and neuronal growth stimulus (90) in animal models. Since neuroinflammation is implicated in NDs that demonstrate memory and cognitive impairment, reports of anti-inflammatory effects could also be of relevance. This is indeed suggested, for instance, for *Vaccinium* spp, (in which there is a report of anti-inflammatory ethnological use (125) there are human studies confirming both memory (126) and cognitive improvement (127). This raises the possibility that the anti-inflammatory effects reported in the ethnological use contribute to the memory and cognitive improvements.

In addition to bioactivity reports, multiple reports of therapeutic benefit from a particular species could also reinforce the fidelity of the benefit. For instance, for *Centella asiatica*, memory improvement effects were reported from surveys of ethnological peoples residing on different continents, in Malaysia (122) and Fiji (121). *Morus alba* has an anti-hypertensive ethnological use reported in China (128) and Trinidad in the Caribbean (129). Other species have multiple reports of memory improvement from within a country, such as for *Musa* × *paradisiaca* within Nigeria (130) (131) and the two reports for *Moringa oleifera* within Kenya (132).

Another measure to aid confirmation of validity of a use could be the number of informant reports per pathological category per species. The rationale for this is that commonly used plants could be more likely to be pharmacologically active (133). Such information was stated in only a few of the studies. For instance, in a Spanish survey 22 informants reported anti-microbial effects for *Thymus vulgaris* (134); and with the Hakka people of China there were 9 reports of anti-inflammatory effects for *Pholidota chinensis* (135). Some studies (e.g. (136) stated informant numbers for each species without reference to the number of informants for each disease category for that species, which was of limited use. More commonly, studies employed measures of consensus such as the use value (UV) (defined as the number of citations per species divided by the number of informants) (e.g. as used by (137)). However, such studies tend to calculate multiple reported uses for any species, including uses not relevant to NDs, precluding the application of such measures to assess validity of therapeutic effects only specific to NDs. An exception is if the requirement is to

find plants with many uses, indicated by plants with higher UVs (for examples see (138)) which is also desirable if the uses are of ND relevance. A useful ethnomedical study in Kenya helpfully listed both numbers of informants for each ailment for each species, alongside analyses such as UVs and fidelity levels (FL) (the most useful species used for a particular medical condition). High FL levels for a species (e.g. *Cajanus cajan*) indicated practitioners' outstanding preference for this species to treat a major ailment (139).

Surveying the community's response to the efficacy of traditional medicine could also help in the assessment of validity. For instance, in a survey of 150 people in the High Atlas of Morocco, 45 % of the surveyed found the use of traditional remedies lead to a total cure, 55 % felt the remedies improved health, and 0.49 % found them ineffective (140). It would have been useful if the survey had been extended to discover the degree of effectiveness of cure reported by respondents for each species, as there is a paucity of such studies in the literature.

There is a yet further means by which the validation of plant ethnomedical uses can be explored. This is hinted at in a number of species that have sister species (another species within the same genus) sharing ethnomedical uses in common. For instance, *Cymbopogon citratus, and* the sister species *Cymbopogon giganteus* both share memory improvement use (Supplementary Table 2). *Asparagus africanus* and *Asparagus racemosus* both have a reported anti-paralytic ethnological use. This suggests that such species share a common molecular machinery, as one might expect since there is evidence that members of the same genera are fundamentally related in some ways (141). Moreover, sister species may have additional reported uses of ND therapeutic potential which could complement one another. For instance, there is a reported anti-paralytic use (of relevance to ALS and MS) in *Ardisia gigantifolia. Ardisia crenata* has a reported anti-inflammatory and anti-microbial use which is unreported in *A. gigantifolia*, but since it is a sister species of *A. crenata* it may have this similar capability as a hidden potential, and indeed which may be contributing to the therapeutic effect, particularly since inflammation is implicated in ALS and MS pathology.

Similarly, *Crinum nubicum* has an anti-paralytic reported use, and the sister species *Crinum asiaticum* is reported to neutralise poisons, suggesting that the Crinum genus may have anti-neurotoxic properties which could be promising for further investigation.

37 out of 47 (79 %) of these species clusters sharing a common therapeutic use have an intercontinental distribution, which may give further weight to the validity of the reports.

## 5. MULTIPLE THERAPEUTIC USES

Species with medicinal uses for some of these categories are of simultaneous relevance to more than one ND, and thus have a potentially wide therapeutic scope. For instance, the species with reported memory/cognitive improvement (summarized in Table 2) are of relevance not only to AD but also A-H, BD, CS, FRDA, HD, MSA, MS, NPC, PD and VD, since all of these diseases manifest memory/cognitive impairment.

The results indicate that species with reports of ND potential from multiple therapeutic categories are common. 734 of the 2001 species (37 %) demonstrated more than one therapeutic use of ND potential (Supplementary Table 1). 249 species (12 %) demonstrated at least three reported such uses. 48 species demonstrated at least five uses (Table 5).

**Table 5** Plant species with at least five reported ethnological uses of ND potential

| Species | Reported ethnomedical uses |
|---|---|
| *Achyranthes aspera* | Anti-paralytic; anti-inflammatory; anti-microbial; anti-epileptic; wound healing |
| *Aframomum melegueta* | Hemiplegia/ paraplegia/ polio; memory improvement/ enhancement; anti-microbial; stroke; TBI |
| *Allium sativum* | Anti-hypertensive; anti-venom; anti-inflammatory; anti-viral; anti-paralytic/polio; memory improvement/ enhancement; anti-epileptic; immunostimulant; skin regeneration; anti-venom; anti-stroke |
| *Alstonia scholaris* | Anti-paralytic; cerebral palsy; anti-inflammatory; anti-viral; anti-hypertensive; anti-venom |
| *Annona muricata* | Anti-hypertensive; anti-inflammatory; anti-microbial;; anti-AD, anti-dementia; TBI |
| *Bixa orellana* | Anti-inflammatory; anti-microbial; anti-hypertensive; cholesterol-lowering; anti-venom; , |
| *Blumea balsamifera* | Limb paralysis, muscle spasms; anti-inflammatory; anti-microbial; anti-hypertensive |
| *Boerhavia diffusa* | Anti-paralytic, stimulates new tissue in wound healing; anti-venom; anti-microbial; anti-inflammatory; neurological disorders |
| *Borago officinalis* | Anti-microbial; anti-hypertensive; hypercholesterolemia; anxiolytic; anti-inflammatory; memory improvement |
| *Cajanus cajan* | Anti-microbial, thermal shock; anti-inflammatory; anti-hypertensive; anti-venom; anti-epileptic |
| *Calotropis procera* | Anti-bacterial; anti-oxidant; antifungal, anti-viral; anti-paralytic; anti-venom; wound healing; anti-inflammatory; epilepsy; anti-stroke, anti-PD |
| *Carica papaya* | Memory enhancement; anti-oxidant, anti-hypertensive; anti-microbial; boost immune system; anti-inflammatory; wound healing |
| *Cassia fistula* | Anti-paralytic; anti-microbial; anti-venom; anti-epileptic; anti-inflammatory |
| *Centella asiatica* | Memory improvement; brain tonic; leg weakness; anti-inflammatory; anti-microbial; anti-venom; anti-hypertensive |
| *Dipteryx alata* | Anti-microbial; anti-inflammatory; cholesterol-reducing; anti-thrombotic; memory improvement; anti-venom |

| | |
|---|---|
| *Dysphania ambrosioides* | Anti-microbial; memory improvement; anti-inflammatory; anti-hypertensive; anti-stroke |
| *Jatropha curcas* | Anti-paralytic; anti-microbial; memory improvement/ enhancement; anti-convulsant; mental disorder; anti-inflammatory; anti-venom; wound healing; anti-toxin |
| *Kalanchoe pinnata* | Anti-viral; anti-aging; anti-hypertensive; anti-inflammatory; anti-paralytic |
| *Lippia multiflora* | Anti-hypertensive; anti-microbial; anti-AD; anti-PD; epilepsy; stroke |
| *Mangifera indica* | Anti-microbial; anti-inflammatory; anti-hypertensive; memory improvement/ enhancement; boost immune system; wound healing |
| *Melissa officinalis* | Anti-hypertensive; anti-inflammatory; anti-microbial; anxiolytic; memory improvement |
| *Mentha spicata* | Anti-microbial; anti-viral; anti-hypertensive; anti-inflammatory; fatigue; anxiolytic |
| *Moringa oleifera* | Anti-microbial; anti-inflammatory; anti-microbial/anti-paralytic; memory enhancement; fatigue; anti-epileptic; wound healing |
| *Mucuna pruriens* | Anti-microbial; anti-paralytic; tissue regeneration; anti-epileptic; anti-PD; anti-venom |
| *Nauclea latifolia* | Anti-paralytic/polio; memory improvement; anti-inflammatory; anti-microbial; TBI |
| *Ocimum tenuiflorum* | Anti-inflammatory; anti-microbial; enhanced memory; anti-paralytic; wound healing |
| *Origanum vulgare* | Anti-microbial; anti-inflammatory; anti-paralytic; anti-hypertensive; fatigue, weakness |
| *Oroxylum indicum* | Anti-microbial; anti-hypertensive; anti-inflammatory , rheum; anti-venom; anxiolytic |
| *Peperomia pellucida* | Anti-inflammatory; anti-microbial; anti-hypertensive; anti-aging; anti-venom |
| *Phyllanthus amarus* | Anti-paralytic; anti-hypertensive; anti-inflammatory; anti-microbial; memory improvement/ enhancement |
| *Phyllanthus emblica* | Anti-hypertensive; anti-microbial; anti-aging; anti-inflammatory; anti-fatigue |
| *Plantago major* | neutralize venom; anti-microbial; anti-inflammatory; anti-hypertensive; wound healing |
| *Plumbago zeylanica* | Anti-microbial; memory improvement; anti-paralytic/polio; anti-venom; anti-inflammatory |
| *Polygonatum sibiricum* | Support immune system, anti-aging; anti-bacterial; anti-hypertensive; anti-fatigue |
| *Psidium guajava* | Anti-microbial; anti-inflammatory; anti-epileptic; wound; anxiolytic |
| *Rosmarinus officinalis* | Anti-microbial; anti-inflammatory; wound healing; anti-atherosclerotic; memory; anti-hypercholesterol; fatigue; anti-hypertensive |
| *Salvadora persica* | Anti-microbial; poison antidote, anti-venom; low immunity; anti-inflammatory |
| *Securidaca longipedunculata* | Anti-PD; anti-inflammatory; anti-microbial; dementia, memory, epilepsy |
| *Scoparia dulcis* | Memory enhancement; anti-microbial; anti-inflammatory; anti-hypertensive; anti-venom |
| *Senna occidentalis* | Anti-inflammatory; anti-hypertensive; anti-microbial; memory improvement/ enhancement; anti-convulsant |
| *Sesbania sesban* | Anti-microbial; anti-inflammatory; anti-venom; promote new tissue formation; anti-hypertensive |
| *Sida acuta* | Anti-inflammatory; anti-microbial; anti-paralytic; anti-venom; nervous diseases |
| *Solanum indicum* | Anti-microbial; boost energy; anti-inflammatory; boost immune system; anti-venom |
| *Tamarindus indica* | Anti-venom; anti-microbial; anti-hypertensive; memory improvement; anti-inflammatory |
| *Urtica dioica* | Anti-paralytic; anti-microbial; anti-hypertensive; anxiolytic; anti-inflammatory |
| *Vitex negundo* | Anti-microbial; anti-inflammatory; anti-epileptic; memory improvement; anti-venom |
| *Withania somnifera* | Anti-microbial; anti-venom; anti-inflammatory; wound healing; anti-paralytic; poison antidote |
| *Zingiber officinale* | Anti-microbial; anti-inflammatory; memory improvement/enhancement; wound healing; boost immune system, anti-fatigue; anti-paralytic |

The phenomenon of multiple medicinal effects of certain individual plant species is well recognized (142), and this is found to be the case here too for plants with ND therapeutic potential. This is highly relevant for NDs, in which several hallmarks of pathology may act together interdependently.

As a result, it is increasingly recognised that the failure of therapies aimed at reversing these diseases may be attributed to this complexity, and is a reason why single drugs aimed at just one pathological target have failed (1). Researchers are therefore looking at drugs aimed at these multiple pathological targets (143) (144) (145), so the plant species with reports of multiple effects on ND pathologies could be of great therapeutic potential.

In many species demonstrating multiple therapeutic effects, anti-inflammatory benefit is one of the most common. For instance, in both *Securidaca longipedunculata* and *Cecropia pachystachya* the dual effects of anti-PD and anti-inflammation are reported, which is of relevance to PD and MSA. Anti-inflammatory effects are prominent for almost all of the species showing five or more therapeutic effects (Table 5), with anti-inflammatory benefit reported in 44 of the 48 species (92 %). Since inflammation is implicated in many if not all NDs and may even be the driver, neuroinflammatory processes could be a therapeutic target with these species, but with the capability to simultaneously target other key ND pathologies.

Table 5 gives numerous examples of multiple uses that could therapeutically act together. For instance, *Dysphania ambrosioides,* is reported to confer memory improvement, anti-hypertensive and anti-stroke therapeutic effects. Similarly, *Rosmarinus officinalis* is reported to confer memory improvement, anti-atherosclerotic and anti-hypercholesterol effects. Together these effects are all relevant to AD and vascular dementia, since cardiovascular pathology is implicated in these diseases. Anti-paralytic, anti-venom, anti-toxin and anti-inflammatory effects are reported for both *Jatropha curcas and Allium sativum*, which could all be of therapeutic relevance for ALS and MS. Additionally *A. sativum* has reported anti-HIV effects, which is of relevance since retroviruses have been implicated in ALS etiology. Anti-paralytic, tissue regeneration wound healing and anti-venom effects reported in *Mucuna pruriens* is also of potential relevance to ALS and MS. Anti-venom and anti-toxin uses could be targeted to the neurotoxic factors driving these diseases.

It is common to find species in which multiple ethnological uses are mirrored by the same species demonstrating multiple bioactivities of ND relevance. For instance, in *Moringa oleifera* the ethnological reports of beneficial effects of memory improvement, anti-inflammatory and anti-paralytic beneficial effects are mirrored or confirmed by bioactivities including memory

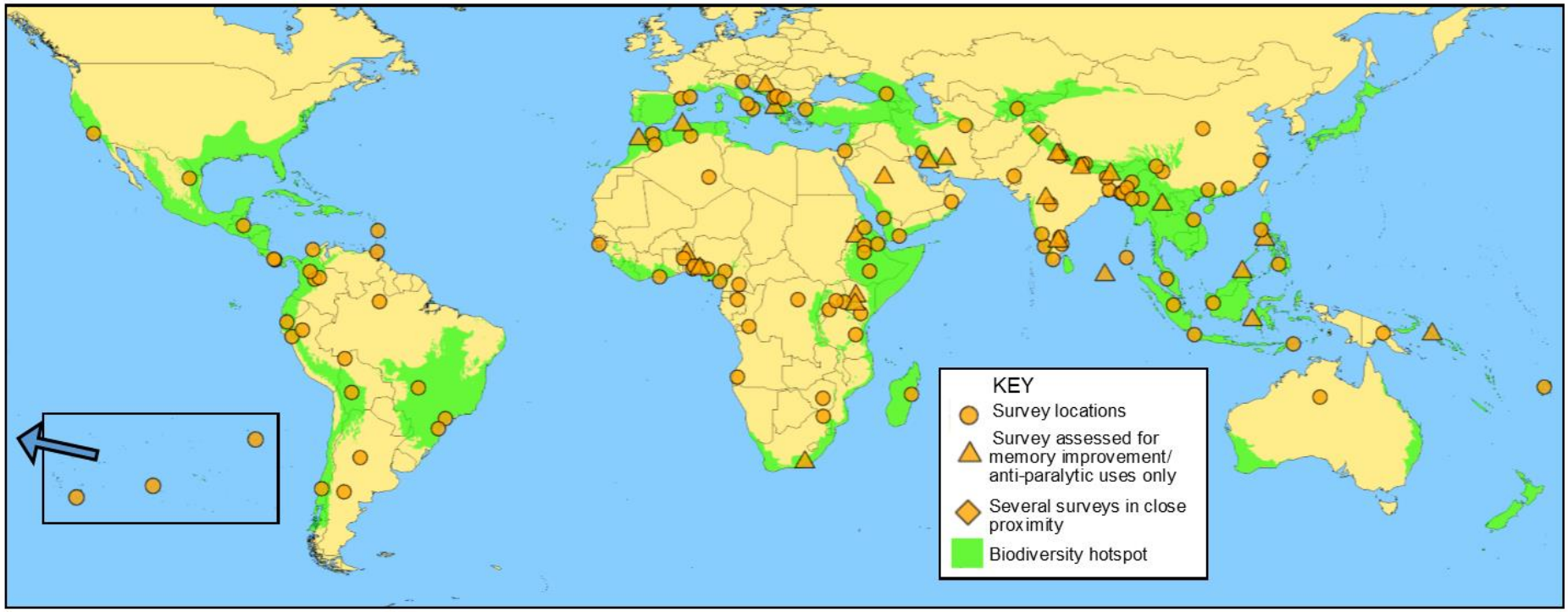
improvement, anti-inflammatory and delayed ALS disease phenotypes. These multiple activities confirm the ethnological uses and suggest that the multiple effects of such plants is of relevance to target the pluralistic pathologies of NDs.

## 6. DISTRIBUTION OF SURVEYS

### *6.1 Overall distribution pattern of surveys with ND relevance*

The distribution of the surveys from which data of ND relevance was mined are indicated in Figure 1. Of the 67 countries represented by the surveys, these were located most commonly in three continents: Africa 19 surveys; Asia 20 surveys; and the Americas (South, Central and North America combined) 13 surveys. This suggests a particular abundance of studies of ND relevance from those continents. More than one survey within a country was mined if there were sufficiently distinct habitat types or regional identity between them (e.g. Amazonian versus Atlantic Forest of Brazil) or where ND-relevant data richness was revealed in the literature searches. The latter is exemplified by India, in which 10 separate surveys were found that cited species with uses reported for memory improvement. The overall distribution pattern of the surveys across the world revealed by online search engines reflects the abundance of ethnomedical surveys for certain countries (e.g. India, Nigeria), but a paucity of surveys for other countries (e.g. Chad, Libya). Various search term alternatives were needed to obtain surveys for certain countries (such as some islands of Indonesia), suggesting that some journals have more modest search engine reach.

**Figure 1** Distribution of 157 ethnomedical surveys with potential therapeutic relevance for neurodegenerative diseases.




The survey distribution on the map indicates that the surveys were located most commonly in 3 continents: Africa, Asia and the Americas, suggesting an abundance of studies of ND relevance from those continents. There is also an abundance of surveys for certain countries (e.g. India, Nigeria), with the biggest cluster of surveys (40 in all) in India, Bangladesh and Pakistan combined. 90 out of 157 (57 %) of the studies were found to reside within biodiversity hotspots and therefore are under threat.

Figure 1 demonstrates that the biggest cluster of surveys (40 in total) occurs in India, Bangladesh and Pakistan. This cluster is concentrated particularly in the Himalayan ranges and environs, and although this may be associated with the very high species diversity there, there are other countries with even higher species diversity in which ethnomedical surveys are less common, such as parts of South America. For instance, the Pacific region of Columbia has one of the highest pockets of biodiversity in the world with 50,000 species of plants, and yet both scientific knowledge on Colombian flora and ethnomedical investigations are lacking (146). The cluster of surveys yielding such high levels of data in the India-Bangladesh-Pakistan country block could be attributed to the philosophy of the Ayurvedic medicine system, in which every plant on earth is considered to have a medicinal property (147), which provides great motivation to search for novel medicinal plant uses, although this could return false positives.

The distribution of studies reporting therapeutic use for key pathologies relevant to NDs was ascertained using memory improvement and anti-paralytic benefits as examples (Figure 1). Studies located in Africa provide the largest concentration of reports for memory improvement, with 95 species out of a total number of 130 (73 %) being found there. The reasons for this are not easy to discern, but there could be a number of contributory factors. 45 of these species were found from Prelude (the African online database) (29), but whilst this assisted in discovery of the data it still does not explain the richness of the data within the African continent. Four of the published ethnomedical surveys focussed specifically on memory improvement (131) (148) (149) in Nigeria and Ethiopia, and central nervous system disorders including memory loss and AD in Togo (150). These publications would reflect a study design in which a specific aim of the authors would have been to search for reports of memory improvement in the ethnomedical community. However, such studies were only a small number out of the 59 published surveys reporting species that confer memory improvement. This is a considerable number of publications, giving some weight to the validity of ethnomedical use to remediate memory loss pathologies.

Another serious pathology common to NDs such as ALS is paralysis. The 185 species with anti-paralytic effects were reported in studies located in all inhabited continents, but most commonly in

Africa with 67 species (36 %), Asia with 88 species (48 %) and 22 species (12 %) in the Americas. Two of the published papers ((151) in the Middle East and (152) in India) focussed specifically on anti-paralytic uses, but this is a small number of studies in relation to the 85 publications reporting species with anti-paralytic benefit. This again is a considerable number of publications, giving some weight to the validity of ethnomedical use to remediate paralysis.

However, even focuses specific to serious particular pathologies such as memory loss or paralysis do not completely explain the abundance of this data in the communities studied in these continents, but merely facilitate the revealing of the data possessed by the communities. One possibility may be that the data richness is associated with a relatively high level of importance of ethnomedical plant use to the community, which is examined in more detail as follows.

### *6.2 Factors affecting ND therapeutic importance tend to be higher in regions where ethnomedical plant use is vital to the community*

An assessment of ND therapeutic potential of a selection of the surveys was made by determining plant uses for the range of serious diseases and pathologies (RSD), defined as causing adverse, life-changing effects or death. In general, the studies located in regions where ethnomedical plant use is vital to the health of the community exhibited plant uses with a high RSD (Supplementary Table 3). This is shown, for instance, in studies within Indian states of Mizoram (with an RSD of ND relevance of 8, and RSD of 10 for all serious pathologies) (denoted as 8 [10]) and Tamil Nadu (7 [8]); and the Maonan people of China (9 [10]). Conversely, studies located in regions in which ethnomedical plant use is not important to the majority of the population's health demonstrated a low RSD. This is exemplified by two studies in Italy (both 0 [0]).

The association between the degree of importance of ethnomedicine to the community and the RSD is to be expected, since if ethnomedicine is vital, it is the driver to search for remedies for serious diseases and pathologies and preserve the knowledge of such remedies. At the same time communities with higher RSDs need to be preserved from the issues that have driven down the RSDs in countries such as Italy and Spain and threaten to do so globally.

However, the RSD reported in a study can be influenced by a number of factors including the number of informants, timescale of the study, the level of informants' expertise, location size, habitat biodiversity, questionnaire design, co-operation of the indigenous people (vs. keeping their ethnomedical knowledge secret) and sociological effects such as war and migration.

The study within East Timor exemplifies low informant number and short study duration: only one traditional medicine expert was made available by local authorities, and the consultation lasted only eight days (153). However, this same study provides an invaluable insight into the use of medicinal plants in a war conflict situation. In this case, resistance workers fled from the occupying Indonesian forces into remote jungle regions, in which a knowledge of useful forest plants became essential to the resistance soldiers' survival. The war situation influenced the soldiers' medical plant needs, such as for species providing therapeutic effects for serious life threatening wounds. Moreover, this study provides vital data for Timor, since it was the only ethnomedical survey for Timor revealed by the online searches. Finally, the timescale of studies ranged from eight days (the Timor survey) to 26 years for a survey in Argentina (154). The collection of more data was in general associated with longer duration of studies and the number of informants questioned.

### *6.3 Data at risk - an assessment of causes of concern*

The surveys were then mapped in relation to the locations of biodiversity hotspots (Figure 1). 90 out of 157 (57 %) of the studies were found to reside within biodiversity hotspots and therefore are under threat. Since BDH regions have by definition lost at least 70 % of their native vegetation, species with valuable therapeutic potential may already have been lost, and there is a threat to the survival of the remaining species. Many of the species listed in the Supplementary Table 1 are not restricted to these regions, but do include numerous endemics or rare/endangered species too. For instance, in Myanmar (which is almost all designated as a BDH) *Aquilaria malaccensis,* which has reported anti-venom, anti-paralytic and anti-viral properties, is critically endangered. The threats are not limited to the BDH regions either. The BDH regions represent only 36 of the world's most threatened areas, with numerous areas outside BDH status still being vulnerable, such as much of the Brazilian Amazon forest (155). Another example

outside a BDH is a study in Mizoram, India, in which 13 of the 81 species of therapeutic ND relevance listed were reported to be rare, vulnerable or endangered there (156).

Next the 115 main surveys were examined systematically to determine what the specific threats of concern were to the authors. The threat of most common concern was loss of traditional knowledge (reported by 58 % of authors). This was a problem reported in communities worldwide, ranging from South America to Europe, Asia and the Pacific. For instance, in Albania the knowledge erosion is due to urbanization and economic migration away from the villages (157). In Saudi Arabia, rapid urbanisation is threatening the very existence of rural communities and their knowledge of medicinal plants (158). In Guangdong, China, the experience of Hakka herbalists gained over generations could be lost due to the disappearance of the traditional culture (135). In Ethiopia, India and Nepal the knowledge erosion is attributed to the younger generation's disinterest in ethnomedical knowledge or a lack of knowledge flow (159) (160) (161). In Fiji, Martinique in the Caribbean and the Philippines its decline is the result of preferences for Western-type medicine (121) (162) (163). Once such knowledge is lost, a major consequence reported in Italy is that the remedies that remain treat only unimportant pathologies (164).

The second-most common threat was habitat loss (reported by 34 % of authors). For instance, the Atlantic Forest region of Brazil agriculture is based on brazilwood, sugarcane, coffee and cattle; along with relocating 50 % of the Brazilian population to cities once covered by forest resulted in only 5 % of the original forest remaining (165). In Ethiopia, habitat loss resulted from various anthropogenic threats such as deforestation to expand agricultural land and firewood collection, according to Getaneh and colleagues(161), who also cite other studies reporting frequent fire, harvesting medicinal plants for building construction and drought (166) (167). Medicinal plants can also be under threat from invasive weeds (168) and grazing (169). Over-harvesting is an issue in countries such as Peru (170). This can lead to species with ND therapeutic potential becoming rare, as for *Sideritis athoa* in Turkey (169) and in Vietnam for *Aquilaria crassna*, which is now critically endangered (171).

Loss of medicinal knowledge and habitat loss are often intertwined. As Ji and colleagues comment regarding the Lisu people, NW Yunnan, China, over-exploitation and deforestation have led to

disappearance of some medicinal plants and the associated knowledge of their use (172). Similar such associations were reported in Côte d'Ivoire (138) and Ethiopia (168).

Whilst there is a need for the protection of medicinal plants from over-harvesting and other threats to their loss, protection can also result in knowledge loss. This is apparent in the ethnomedical survey of the Três Ladeiras community at a forest reserve within Brazil's Atlantic Forest (165). The reserve is protected, so plant collection is prevented. This forces the local specialists to rely more on weed and cultivated plants, including non-native ones. This is in spite of the great importance of the prohibited plants to the community, and the local herbalists' specialist knowledge of these resources is likely to fade with time. The RSD within Brazil's Mato Grosso region was much greater (14 [19]) than for the Atlantic Forest study (4 [5]) in spite of both studies residing in regions of high biodiversity. This suggests that a high RSD is only likely to be realized where the herbalists have access to the full range of plants of therapeutic potential. Thus ideally ways need to be sought to both conserve plant resources that are indeed endangered, but not at the cost of endangering the valuable indigenous knowledge of such plants. In contrast to the Brazilian Três Ladeiras community barred from collecting in the forest reserve, in the Mabira Central Forest Reserve of Uganda the harvesting of medicinal plants from the forest is permitted. This may explain the higher RSD there of 7 [11] compared with 4 [5] for the Três Ladeiras community.

The Bolivian study (173) shows that even with a modest number of informants (21) a good RSD (6 [9]) is achieved, and is associated with most of the Tacana community being reliant on medicinal plants.

## 7. LIMITATIONS OF THE STUDY

This study was restricted to therapies for symptoms and diseases that could be recognized by ethnomedical practitioners. So, for instance motor neurone disease might not be recognized, but instead only therapeutic effects for paralysis (a key symptom of these diseases). Other rare diseases (e.g. Batten, Friedreich's ataxia) would similarly fail to be recognized. Moreover, whilst

some pathological hallmarks are common to more than one ND, the therapeutic effect reported in the ethnological literature may not discriminate between diseases. Again regarding paralysis for instance, anti-paralytic effects could be therapeutic for non-ND conditions such as spinal cord injuries and strokes. The number of publications and thus the prospective numbers of species with ND therapeutic potential was limited by restriction to literature written in the English language, published online. There is a wealth of further promising data in other languages and in printed form which is not online-accessible. Data for South American publications are predominantly in Spanish or Portuguese. The mining of this data by researchers fluent in these languages would thus hold promise for further insights into the therapeutic potential of this high biodiversity continent. Finally, the exclusion of studies in which species were selected according to validation by bioactivity reports was necessary because this study was specifically designed in conjunction with a forthcoming study to find the extent to which the ethnomedical reports are confirmed by bioactivity reports. A more inclusive study not limited in these ways would yield an even greater wealth of useful data.

## 8. TOXICITY ISSUES

Plants with known serious toxicity, such as *Aconitum carmichaeli,* were removed from the study according to the exclusion criteria. There have been over 45 aconite poisoning cases and 3 fatalities reported *from Aconitum* species (reviewed by174). Long-time decoction is the traditional method of detoxification, but following this process hepatoxicity has still been found to remain (174).

Certain plant species for which toxicity reports do exist are nonetheless very popular for traditional medical use. Such species are not removed from the study, but are instead denoted with an asterisk and toxicity study reference in the Supplementary Table 1. For instance, teratogenic defects were found for *Andrographis paniculata, Curcuma longa* and *Carthamus tinctorius* in the zebrafish embryotoxicity model (reviewed by (175). Toxic effects for *Curcuma longa* were observed at only 62.50 *μ*g/mL and mortality and teratogenic effects were found above 125.0 μg/mL in a zebrafish embryo model (176).

## 9. FUTURE DIRECTIONS

Countries not included in this study which remain to be surveyed, along with publications in foreign languages and in printed form are likely to yield yet more data of therapeutic potential.

The standard of publications was variable. In particular, evidence of correct identification of species was sparse or ambiguous in some surveys, although to maximise inclusiveness of data authors were given the benefit of the doubt in such cases where possible. For instance, collected specimens were referred to as being “deposited in herbaria” without reference to how this aided identification, if indeed it did so. However, some studies demonstrated the opposite, i.e. evidence of accurate identification of plant specimens by botanical specialists, and especially when compared with herbarium voucher specimens. Examples of this commendable methodology include the Ethiopian study by Getaneh and Girma (161) in which specimens were compared with authentic herbarium specimens and finally confirmed by taxonomists. Another good example is the survey of the Marquesas Islands of French Polynesia by Girardi and colleagues (136) in which specimens were identified by a botanist in the survey group and compared to voucher specimens taking into account recent taxonomic revisions. Such reassurances are important to ensure species are identified accurately, particularly for species of high therapeutic potential, so it is to be hoped that future surveys attain to more accurate methodologies of identification.

Analysis of the knowledge erosion problem can be found in the study by Voeks and Leony (177), who attribute the key reason for this loss to formal education access, in which the healing properties of their forests and fields no longer find their way into the curriculum. In contrast to this, traditional knowledge (TK) is sustained in the Kenyan masai tribe by children spending time with their parents, and this TK persists even with children’s enrolment into formal education (178).
It is beyond the scope of this study to discuss in detail the threats of concern to the survey authors. However, several remedial strategies are illustrated from the authors surveyed in this study. For instance, harvesting of medicinal plants which are introduced species can be encouraged because these often have little or no impact on the local habitat, in order to preserve sites of native species

under threat (179). This low-impact harvesting of medicinal plants can bring economic benefits, such as the agro-industrial credit initiative in Panama for producers of medicinal plants that can be marketed (180). In the Hakka communities of China there is already an awareness of which plants are endangered, and over-harvesting is prevented by using more common species (135). With local education initiatives, such attitudes could become more widespread. Environmental education can be fostered by key individuals of a community being included into management programs (179). Another community that is very aware of the need to protect their medicinal plants are the Nicobarese, who harvest mainly the leaves of the plants, since these are the most renewable parts (181). Bussmann and Sharon (170) report in Northern Peru where healers are open to new knowledge, watching international health trends to incorporate new species such as Noni (*Morinda citrifolia*) fruits into their own repertoire, the fruit products being harnessed in local plant pharmacies to benefit the local economy and population.

In certain locations the ethnomedical knowledge appeared to still be buoyant, as suggested by an Algerian study, in which the age group of 31–40 was the most common among herbalists (137) rather than amongst dwindling numbers of elderly custodians of knowledge. One of the most striking examples of an improvement in ethnomedical knowledge in recent years has occurred in Kyrgyzstan. Under the 70 years of Soviet rule traditional medicinal practices in such Central Asian societies were neglected and suppressed, leading to a loss of TK (182). However, in the post-Soviet era there has been a remarkable revival of ancestral TK (183).

## 10. CONCLUDING REMARKS

From the assessment of ND pathology and clinical symptoms, it was found possible to construct a number of therapeutic categories recognized by ethnomedical practitioners. These categories were then applied as an ethnomedical toolkit to mine for plant species with reported therapeutic effects of ND relevance, which were found in 2001 species. This confirms the hypothesis that the plant kingdom may possess a large reservoir of ND potential. This toolkit methodology appears to provide a promising wide reach in the search of this potential. The companion paper tests the

toolkit usefulness by probing the extent to which the ethnomedical uses are validated by bioactivity reports. However, whilst around 46 % of the angiosperm families are represented in this data, the 2,001 species with ND potential are relatively small in relation to the estimated 352,000 species just within the angiosperms alone. Since many of these species still remain to be surveyed for their ND therapeutic potential, it is likely that the plant kingdom has an even greater repository of potential yet to be tapped. What is also potentially attractive about the data is that there is a clear symptomatic benefit in the reported human population.

From the assessment of factors affecting the richness of ND therapeutic potential, ND therapeutic importance tended to be higher in communities where ethnomedical plant use is vital to the community, since where ethnomedicine is vital, this provides the driver in the search for serious disease remedies. The implication is that such communities have a particularly rich wealth of plant therapeutic potential for future research focus.

Such a rich repository holds great promise in the quest for bioactive molecules with the capacity to reverse the key pathologies of neurodegenerative diseases. However, there is the need for another reversal too: a reversal of fortunes for TK and habitat loss, if this potential is to be conserved. Encouraging signs such as the reversal of TK loss in Central Asian countries such as Kyrgystan indicate that this can indeed be possible.

**Abbreviations**

Aβ, amyloid β; AD, Alzheimer's disease; ALD, Alexander disease; α-syn, alpha-synuclein; A-H, Alpers-Huttenlocher syndrome; ALS, Amyotrophic lateral sclerosis; BBB, blood-brain barrier; BD, Batten disease; BDH, biodiversity hotspots; CJD, Creutzfeldt-Jakob disease; CS, Cockayne syndrome; FTD, Frontotemporal dementia; FRDA, Friedreich's ataxia; HD, Huntington's disease; LSD, Lysosomal storage diseases; MSA, Multiple system atrophy; MS, Multiple sclerosis; MSP, Multisystem proteinopathy; ND, neurodegenerative disease; NPC, Neimann-Pick disease type C; PD, Parkinson's disease; PS, Perry syndrome; SMA, Spinal muscular atrophy; SOD1, superoxide dismutase 1; Syn, synonym; TBI, traumatic brain injury; TDP-43, TAR DNA binding protein; TK, traditional knowledge; VD, Vascular dementia.

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**Authors' contributions**

ST designed the study and wrote the manuscript. LT performed the RStudio graphics mapping and contributed to the writing of manuscript. Both authors read and approved the final manuscript.

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**REFERENCES**

1. Yiannopoulou KG, Anastasiou AI, Zachariou V, Pelidou SH. Reasons for Failed Trials of Disease-Modifying Treatments for Alzheimer Disease and Their Contribution in Recent Research. Biomedicines. 2019;7(4).
2. Reich DS, Lucchinetti CF, Calabresi PA. Multiple Sclerosis. N Engl J Med. 2018;378(2):169-80.
3. Masrori P, Van Damme P. Amyotrophic lateral sclerosis: a clinical review. Eur J Neurol. 2020;27(10):1918-29.

4. Kumar A, Kumar V, Singh K, Kumar S, Kim YS, Lee YM, et al. Therapeutic Advances for Huntington's Disease. Brain Sci. 2020;10(1).
5. Delatycki MB, Bidichandani SI. Friedreich ataxia- pathogenesis and implications for therapies. Neurobiol Dis. 2019;132:104606.
6. Karikkineth AC, Scheibye-Knudsen M, Fivenson E, Croteau DL, Bohr VA. Cockayne syndrome: Clinical features, model systems and pathways. Ageing Res Rev. 2017;33:3-17.
7. Robberecht W, Philips T. The changing scene of amyotrophic lateral sclerosis. Nat Rev Neurosci. 2013;14(4):248-64.
8. Nichols E, Szoeke CE, Vollset SE, Abbasi N, Abd-Allah F, Abdela J, et al. Global, regional, and national burden of Alzheimer's disease and other dementias, 1990–2016: a systematic analysis for the Global Burden of Disease Study 2016. The Lancet Neurology. 2019;18(1):88-106.
9. Fouka M, Mavroeidi P, Tsaka G, Xilouri M. In search of effective treatments targeting α-synuclein toxicity in synucleinopathies: Pros and cons. Frontiers in cell and developmental biology. 2020;8:894.
10. Johnson TB, Cain JT, White KA, Ramirez-Montealegre D, Pearce DA, Weimer JM. Therapeutic landscape for Batten disease: current treatments and future prospects. Nat Rev Neurol. 2019;15(3):161-78.
11. Newman DJ, Cragg GM. Natural products as sources of new drugs over the 30 years from 1981 to 2010. J Nat Prod. 2012;75(3):311-35.
12. Newman DJ, Cragg GM. Natural Products as Sources of New Drugs over the Nearly Four Decades from 01/1981 to 09/2019. J Nat Prod. 2020;83(3):770-803.
13. Tu Y. Artemisinin-A Gift from Traditional Chinese Medicine to the World (Nobel Lecture). Angew Chem Int Ed Engl. 2016;55(35):10210-26.
14. Johnson IS, Armstrong JG, Gorman M, Burnett JP. The vinca alkaloids: a new class of oncolytic agents. Cancer Res. 1963;23:1390-427.
15. Nejat N, Valdiani A, Cahill D, Tan YH, Maziah M, Abiri R. Ornamental exterior versus therapeutic interior of Madagascar periwinkle (Catharanthus roseus): the two faces of a versatile herb. ScientificWorldJournal. 2015;2015:982412.
16. Kew Plant List [Internet]. Available from: http://www.theplantlist.org/.
17. Verpoorte R. Exploration of nature's chemodiversity: the role of secondary metabolites as leads in drug development. Drug Discovery Today. 1998;3(5):232-8.
18. Verpoorte R. Pharmacognosy in the new millennium: leadfinding and biotechnology. J Pharm Pharmacol. 2000;52(3):253-62.
19. Fabricant DS, Farnsworth NR. The value of plants used in traditional medicine for drug discovery. Environ Health Perspect. 2001;109 Suppl 1:69-75.
20. Atanasov AG, Waltenberger B, Pferschy-Wenzig EM, Linder T, Wawrosch C, Uhrin P, et al. Discovery and resupply of pharmacologically active plant-derived natural products: A review. Biotechnol Adv. 2015;33(8):1582-614.
21. Henkin JM, Sydara K, Xayvue M, Souliya O, Kinghorn AD, Burdette JE, et al. Revisiting the linkage between ethnomedical use and development of new medicines: A novel plant collection strategy towards the discovery of anticancer agents. J Med Plant Res. 2017;11(40):621-34.
22. Adams M, Gmünder F, Hamburger M. Plants traditionally used in age related brain disorders--a survey of ethnobotanical literature. J Ethnopharmacol. 2007;113(3):363-81.
23. Akram M, Nawaz A. Effects of medicinal plants on Alzheimer's disease and memory deficits. Neural Regen Res. 2017;12(4):660-70.
24. Howes MJ, Houghton PJ. Ethnobotanical treatment strategies against Alzheimer's disease. Curr Alzheimer Res. 2012;9(1):67-85.
25. Javed H, Nagoor Meeran MF, Azimullah S, Adem A, Sadek B, Ojha SK. Plant extracts and phytochemicals targeting α-synuclein aggregation in Parkinson's disease models. Frontiers in pharmacology. 2019;9:1555.
26. Myers N, Mittermeier RA, Mittermeier CG, da Fonseca GA, Kent J. Biodiversity hotspots for conservation priorities. Nature. 2000;403(6772):853-8.
27. Mittermeier RA, Turner WR, Larsen FW, Brooks TM, Gascon C. Global biodiversity conservation: the critical role of hotspots. Biodiversity hotspots: Springer; 2011. p. 3-22.
28. Moher D, Liberati A, Tetzlaff J, Altman DG, Group P. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. PLoS Med. 2009;6(7):e1000097.

29. Noe N, Lehmann J. Prelude medicinal plants. Belgian Biodiversity Platform. Database. 2012.
30. Native American Ethnobotany [Internet]. [cited 2.9.2021]. Available from: http://naeb.brit.org/.

31. Kew science online medicinal-plant-names-services [Internet]. Royal Botanic Gardens, Kew. Available from: https://mpns.science.kew.org/
32. World Flora Online [Internet]. William Ulate. Available from: http://www.worldfloraonline.org/.
33. Plants of the World Online [Internet]. Royal Botanic Gardens, Kew Available from: http://www.plantsoftheworldonline.org/.
34. Ribeiro RV, Bieski IGC, Balogun SO, Martins DTO. Ethnobotanical study of medicinal plants used by Ribeirinhos in the North Araguaia microregion, Mato Grosso, Brazil. J Ethnopharmacol. 2017;205:69-102.
35. RStudio team: Integrated Development for R. . Boston, MA RStudio; 2020. http://www.rstudio.com

36. Hoffman M, Koenig K, Bunting G, Costanza J, Williams KJ. Biodiversity Hotspots (version 2016.1). 2016.
37. Latlong coordinates reference for World Geodetic System WGS84 standard. [Internet]. Available from: www.latlong.net.
38. Sweeney P, Park H, Baumann M, Dunlop J, Frydman J, Kopito R, et al. Protein misfolding in neurodegenerative diseases: implications and strategies. Transl Neurodegener. 2017;6:6.
39. Boland B, Yu WH, Corti O, Mollereau B, Henriques A, Bezard E, et al. Promoting the clearance of neurotoxic proteins in neurodegenerative disorders of ageing. Nat Rev Drug Discov. 2018;17(9):660-88.
40. Heneka MT, Carson MJ, El Khoury J, Landreth GE, Brosseron F, Feinstein DL, et al. Neuroinflammation in Alzheimer's disease. Lancet Neurol. 2015;14(4):388-405.
41. Arlt S. Non-Alzheimer's disease-related memory impairment and dementia. Dialogues Clin Neurosci. 2013;15(4):465-73.
42. Trapp BD, Nave KA. Multiple sclerosis: an immune or neurodegenerative disorder? Annu Rev Neurosci. 2008;31:247-69.
43. La Rosa P, Bertini ES, Piemonte F. The NRF2 Signaling Network Defines Clinical Biomarkers and Therapeutic Opportunity in Friedreich's Ataxia. Int J Mol Sci. 2020;21(3).
44. Li Hi Shing S, Chipika RH, Finegan E, Murray D, Hardiman O, Bede P. Post-polio Syndrome: More Than Just a Lower Motor Neuron Disease. Front Neurol. 2019;10:773.
45. Wirdefeldt K, Adami H-O, Cole P, Trichopoulos D, Mandel J. Epidemiology and etiology of Parkinson's disease: a review of the evidence. European journal of epidemiology. 2011;26(1):1.
46. Guzman-Martinez L, Maccioni RB, Andrade V, Navarrete LP, Pastor MG, Ramos-Escobar N. Neuroinflammation as a Common Feature of Neurodegenerative Disorders. Front Pharmacol. 2019;10:1008.
47. Hickman S, Izzy S, Sen P, Morsett L, El Khoury J. Microglia in neurodegeneration. Nat Neurosci. 2018;21(10):1359-69.
48. Mu Y, Gage FH. Adult hippocampal neurogenesis and its role in Alzheimer's disease. Mol Neurodegener. 2011;6:85.
49. Hammond TR, Marsh SE, Stevens B. Immune Signaling in Neurodegeneration. Immunity. 2019;50(4):955-74.
50. Winner B, Winkler J. Adult neurogenesis in neurodegenerative diseases. Cold Spring Harb Perspect Biol. 2015;7(4):a021287.
51. Monzio Compagnoni G, Di Fonzo A. Understanding the pathogenesis of multiple system atrophy: state of the art and future perspectives. Acta Neuropathol Commun. 2019;7(1):113.
52. Saffari A, Kölker S, Hoffmann GF, Ebrahimi-Fakhari D. Linking mitochondrial dysfunction to neurodegeneration in lysosomal storage diseases. J Inherit Metab Dis. 2017;40(5):631-40.
53. Brunetti-Pierri N, Scaglia F. GM1 gangliosidosis: review of clinical, molecular, and therapeutic aspects. Mol Genet Metab. 2008;94(4):391-6.
54. Morena J, Gupta A, Hoyle JC. Charcot-Marie-Tooth: From Molecules to Therapy. Int J Mol Sci. 2019;20(14).

55. Rahman S, Copeland WC. POLG-related disorders and their neurological manifestations. Nat Rev Neurol. 2019;15(1):40-52.
56. Itzhaki RF, Lathe R, Balin BJ, Ball MJ, Bearer EL, Braak H, et al. Microbes and Alzheimer's Disease. J Alzheimers Dis. 2016;51(4):979-84.
57. López-Otín C, Blasco MA, Partridge L, Serrano M, Kroemer G. The hallmarks of aging. Cell. 2013;153(6):1194-217.
58. Lathe R, Sapronova A, Kotelevtsev Y. Atherosclerosis and Alzheimer-diseases with a common cause? Inflammation, oxysterols, vasculature. BMC geriatrics. 2014;14(1):1-30.
59. Raz L, Bhaskar K, Weaver J, Marini S, Zhang Q, Thompson JF, et al. Hypoxia promotes tau hyperphosphorylation with associated neuropathology in vascular dysfunction. Neurobiol Dis. 2019;126:124-36.
60. Iadecola C, Yaffe K, Biller J, Bratzke LC, Faraci FM, Gorelick PB, et al. Impact of Hypertension on Cognitive Function: A Scientific Statement From the American Heart Association. Hypertension. 2016;68(6):e67-e94.
61. Bräuer AU, Kuhla A, Holzmann C, Wree A, Witt M. Current Challenges in Understanding the Cellular and Molecular Mechanisms in Niemann-Pick Disease Type C1. Int J Mol Sci. 2019;20(18).
62. Toledano-Zaragoza A, Ledesma MD. Addressing neurodegeneration in lysosomal storage disorders: Advances in Niemann Pick diseases. Neuropharmacology. 2020;171:107851.
63. Mendes FR, Negri G, Duarte-Almeida JM, Tabach R, Carlini EA. The action of plants and their constituents on the central nervous system. Plant bioactives and drug discovery: principles, practice, and perspectives. 2012;17:161.
64. Tailor B, Pilozzi A, Huang X. Contributing Factors of Neurodegeneration in Alzheimer's Disease. Exon Publications. 2019:69-84.
65. Streit WJ, Walter SA, Pennell NA. Reactive microgliosis. Prog Neurobiol. 1999;57(6):563-81.
66. Streit WJ, Mrak RE, Griffin WS. Microglia and neuroinflammation: a pathological perspective. J Neuroinflammation. 2004;1(1):14.
67. Burda JE, Sofroniew MV. Reactive gliosis and the multicellular response to CNS damage and disease. Neuron. 2014;81(2):229-48.
68. Jeong S. Molecular and Cellular Basis of Neurodegeneration in Alzheimer's Disease. Mol Cells. 2017;40(9):613-20.
69. Hardiman O, Al-Chalabi A, Chio A, Corr EM, Logroscino G, Robberecht W, et al. Amyotrophic lateral sclerosis. Nat Rev Dis Primers. 2017;3:17085.
70. Spillantini MG, Crowther RA, Jakes R, Cairns NJ, Lantos PL, Goedert M. Filamentous alpha-synuclein inclusions link multiple system atrophy with Parkinson's disease and dementia with Lewy bodies. Neurosci Lett. 1998;251(3):205-8.
71. Kamble N, Pal PK. Tremor syndromes: A review. Neurol India. 2018;66(Supplement):S36-S47.
72. Lecker SH, Goldberg AL, Mitch WE. Protein degradation by the ubiquitin-proteasome pathway in normal and disease states. J Am Soc Nephrol. 2006;17(7):1807-19.
73. Dinarello CA. Proinflammatory cytokines. Chest. 2000;118(2):503-8.
74. Wood LB, Winslow AR, Proctor EA, McGuone D, Mordes DA, Frosch MP, et al. Identification of neurotoxic cytokines by profiling Alzheimer's disease tissues and neuron culture viability screening. Sci Rep. 2015;5:16622.
75. Zhang JM, An J. Cytokines, inflammation, and pain. Int Anesthesiol Clin. 2007;45(2):27-37.
76. Zheng C, Zhou XW, Wang JZ. The dual roles of cytokines in Alzheimer's disease: update on interleukins, TNF-α, TGF-β and IFN-γ. Transl Neurodegener. 2016;5:7.
77. Masgrau R, Guaza C, Ransohoff RM, Galea E. Should we stop saying 'glia'and 'neuroinflammation'? Trends in molecular medicine. 2017;23(6):486-500.
78. Maragakis NJ, Rothstein JD. Mechanisms of Disease: astrocytes in neurodegenerative disease. Nat Clin Pract Neurol. 2006;2(12):679-89.
79. Liddelow SA, Guttenplan KA, Clarke LE, Bennett FC, Bohlen CJ, Schirmer L, et al. Neurotoxic reactive astrocytes are induced by activated microglia. Nature. 2017;541(7638):481-7.
80. Fischer R, Maier O. Interrelation of oxidative stress and inflammation in neurodegenerative disease: role of TNF. Oxid Med Cell Longev. 2015;2015:610813.

81. Cooper JD, Tarczyluk MA, Nelvagal HR. Towards a new understanding of NCL pathogenesis. Biochim Biophys Acta. 2015;1852(10 Pt B):2256-61.
82. Zindler E, Zipp F. Neuronal injury in chronic CNS inflammation. Best Pract Res Clin Anaesthesiol. 2010;24(4):551-62.
83. Jenkinson ML, Bliss MR, Brain AT, Scott DL. Rheumatoid arthritis and senile dementia of the Alzheimer's type. Br J Rheumatol. 1989;28(1):86-8.
84. Nichols MR, St-Pierre MK, Wendeln AC, Makoni NJ, Gouwens LK, Garrad EC, et al. Inflammatory mechanisms in neurodegeneration. J Neurochem. 2019;149(5):562-81.
85. Czirr E, Wyss-Coray T. The immunology of neurodegeneration. J Clin Invest. 2012;122(4):1156-63.
86. Grote HE, Hannan AJ. Regulators of adult neurogenesis in the healthy and diseased brain. Clin Exp Pharmacol Physiol. 2007;34(5-6):533-45.
87. Altman J, Das GD. Autoradiographic and histological evidence of postnatal hippocampal neurogenesis in rats. J Comp Neurol. 1965;124(3):319-35.
88. Eriksson PS, Perfilieva E, Björk-Eriksson T, Alborn AM, Nordborg C, Peterson DA, et al. Neurogenesis in the adult human hippocampus. Nat Med. 1998;4(11):1313-7.
89. Monje ML, Toda H, Palmer TD. Inflammatory blockade restores adult hippocampal neurogenesis. Science. 2003;302(5651):1760-5.
90. Mohandas Rao KG, Muddanna Rao S, Gurumadhva Rao S. Centella asiatica (L.) leaf extract treatment during the growth spurt period enhances hippocampal CA3 neuronal dendritic arborization in rats. Evid Based Complement Alternat Med. 2006;3(3):349-57.
91. Stys PK, Tsutsui S. Recent advances in understanding multiple sclerosis. F1000Res. 2019;8.
92. Chaudhuri A, Behan PO. Fatigue in neurological disorders. Lancet. 2004;363(9413):978-88.
93. Karshikoff B, Sundelin T, Lasselin J. Role of inflammation in human fatigue: relevance of multidimensional assessments and potential neuronal mechanisms. Frontiers in immunology. 2017;8:21.
94. Gaab J, Rohleder N, Heitz V, Engert V, Schad T, Schürmeyer TH, et al. Stress-induced changes in LPS-induced pro-inflammatory cytokine production in chronic fatigue syndrome. Psychoneuroendocrinology. 2005;30(2):188-98.
95. De Chiara G, Marcocci ME, Sgarbanti R, Civitelli L, Ripoli C, Piacentini R, et al. Infectious agents and neurodegeneration. Mol Neurobiol. 2012;46(3):614-38.
96. Wozniak MA, Frost AL, Preston CM, Itzhaki RF. Antivirals reduce the formation of key Alzheimer's disease molecules in cell cultures acutely infected with herpes simplex virus type 1. PLoS One. 2011;6(10):e25152.
97. Castanedo-Vazquez D, Bosque-Varela P, Sainz-Pelayo A, Riancho J. Infectious agents and amyotrophic lateral sclerosis: another piece of the puzzle of motor neuron degeneration. J Neurol. 2019;266(1):27-36.
98. Niccoli T, Partridge L. Ageing as a risk factor for disease. Curr Biol. 2012;22(17):R741-52.
99. Dzau VJ, Finkelman EM, Balatbat CA, Verdin EM, Pettigrew RI. Achieving healthy human longevity: A global grand challenge. Sci Transl Med. 2020;12(566).
100. Salvadores N, Sanhueza M, Manque P, Court FA. Axonal Degeneration during Aging and Its Functional Role in Neurodegenerative Disorders. Front Neurosci. 2017;11:451.
101. Kalaria RN. Vascular basis for brain degeneration: faltering controls and risk factors for dementia. Nutr Rev. 2010;68 Suppl 2:S74-87.
102. Armstrong RA. Classic β-Amyloid Deposits Cluster Around Large Diameter Blood Vessels Rather than Capillaries in Sporadic Alzheimer's Disease. Current neurovascular research. 2006;3(4):289-94.
103. Roher AE. Cardiovascular system participation in Alzheimer's disease pathogenesis. J Intern Med. 2015;277(4):426-8.
104. Petrovitch H, Ross GW, Steinhorn SC, Abbott RD, Markesbery W, Davis D, et al. AD lesions and infarcts in demented and non-demented Japanese-American men. Ann Neurol. 2005;57(1):98-103.
105. Skoog I, Lernfelt B, Landahl S, Palmertz B, Andreasson LA, Nilsson L, et al. 15-year longitudinal study of blood pressure and dementia. Lancet. 1996;347(9009):1141-5.

106. Iadecola C, Duering M, Hachinski V, Joutel A, Pendlebury ST, Schneider JA, et al. Vascular Cognitive Impairment and Dementia: JACC Scientific Expert Panel. J Am Coll Cardiol. 2019;73(25):3326-44.
107. Langbaum JB, Chen K, Launer LJ, Fleisher AS, Lee W, Liu X, et al. Blood pressure is associated with higher brain amyloid burden and lower glucose metabolism in healthy late middle-age persons. Neurobiol Aging. 2012;33(4):827.e11-9.
108. Aerts JMFG, Kuo CL, Lelieveld LT, Boer DEC, van der Lienden MJC, Overkleeft HS, et al. Glycosphingolipids and lysosomal storage disorders as illustrated by gaucher disease. Curr Opin Chem Biol. 2019;53:204-15.
109. Mole SE, Cotman SL. Genetics of the neuronal ceroid lipofuscinoses (Batten disease). Biochim Biophys Acta. 2015;1852(10 Pt B):2237-41.
110. Radke J, Stenzel W, Goebel HH. Human NCL Neuropathology. Biochim Biophys Acta. 2015;1852(10 Pt B):2262-6.
111. Neugroschl J, Wang S. Alzheimer's disease: diagnosis and treatment across the spectrum of disease severity. Mt Sinai J Med. 2011;78(4):596-612.
112. Ramirez AI, de Hoz R, Salobrar-Garcia E, Salazar JJ, Rojas B, Ajoy D, et al. The Role of Microglia in Retinal Neurodegeneration: Alzheimer's Disease, Parkinson, and Glaucoma. Front Aging Neurosci. 2017;9:214.
113. Adornetto A, Russo R, Parisi V. Neuroinflammation as a target for glaucoma therapy. Neural Regen Res. 2019;14(3):391-4.
114. Caron NS, Dorsey ER, Hayden MR. Therapeutic approaches to Huntington disease: from the bench to the clinic. Nat Rev Drug Discov. 2018;17(10):729-50.
115. Crotti A, Glass CK. The choreography of neuroinflammation in Huntington's disease. Trends Immunol. 2015;36(6):364-73.
116. Alonso R, Pisa D, Carrasco L. Brain Microbiota in Huntington's Disease Patients. Front Microbiol. 2019;10:2622.
117. Jankovic J, Roos RA. Chorea associated with Huntington's disease: to treat or not to treat? Mov Disord. 2014;29(11):1414-8.
118. Navone F, Genevini P, Borgese N. Autophagy and Neurodegeneration: Insights from a Cultured Cell Model of ALS. Cells. 2015;4(3):354-86.
119. Wertz MH, Sahin M. Developing therapies for spinal muscular atrophy. Ann N Y Acad Sci. 2016;1366(1):5-19.
120. Carroll JA, Chesebro B. Neuroinflammation, Microglia, and Cell-Association during Prion Disease. Viruses. 2019;11(1).
121. Singh YN. Traditional medicine in Fiji: some herbal folk cures used by Fiji Indians. J Ethnopharmacol. 1986;15(1):57-88.
122. Awang-Kanak F, Bakar MFA, Mohamed M, editors. Ethnobotanical survey on plants used as traditional salad food (ulam) in Kampung Taun Gusi, Kota Belud Sabah, Malaysia. AIP conference proceedings; 2018: AIP Publishing LLC.
123. Xu MF, Xiong YY, Liu JK, Qian JJ, Zhu L, Gao J. Asiatic acid, a pentacyclic triterpene in Centella asiatica, attenuates glutamate-induced cognitive deficits in mice and apoptosis in SH-SY5Y cells. Acta Pharmacol Sin. 2012;33(5):578-87.
124. Dhanasekaran M, Holcomb LA, Hitt AR, Tharakan B, Porter JW, Young KA, et al. Centella asiatica extract selectively decreases amyloid beta levels in hippocampus of Alzheimer's disease animal model. Phytother Res. 2009;23(1):14-9.
125. Mustafa B, Hajdari A, Pieroni A, Pulaj B, Koro X, Quave CL. A cross-cultural comparison of folk plant uses among Albanians, Bosniaks, Gorani and Turks living in south Kosovo. J Ethnobiol Ethnomed. 2015;11:39.
126. Whyte AR, Cheng N, Fromentin E, Williams CM. A Randomized, Double-Blinded, Placebo-Controlled Study to Compare the Safety and Efficacy of Low Dose Enhanced Wild Blueberry Powder and Wild Blueberry Extract (ThinkBlue™) in Maintenance of Episodic and Working Memory in Older Adults. Nutrients. 2018;10(6).
127. Miller MG, Hamilton DA, Joseph JA, Shukitt-Hale B. Dietary blueberry improves cognition among older adults in a randomized, double-blind, placebo-controlled trial. Eur J Nutr. 2018;57(3):1169-80.

128. Hong L, Guo Z, Huang K, Wei S, Liu B, Meng S, et al. Ethnobotanical study on medicinal plants used by Maonan people in China. Journal of ethnobiology and ethnomedicine. 2015;11(1):32.
129. Clement YN, Baksh-Comeau YS, Seaforth CE. An ethnobotanical survey of medicinal plants in Trinidad. J Ethnobiol Ethnomed. 2015;11:67.
130. Bossard E. La médecine traditionnelle au centre et à l'ouest de l'Angola, 1996. Lisbonne Instituto de Investigaçaö Cientifica Tropical (IICT). 1996;531.
131. Mojisola C, Oladele A, Elufioye T. Ethnobotanical survey of plants used as memory enhancer and antiaging in Ondo State, Nigeria. International Journal of Pharmaceutics. 2012;2:26-32.
132. Kamau LN, Mbaabu PM, Mbaria JM, Gathumbi PK, Kiama SG. Ethnobotanical survey and threats to medicinal plants traditionally used for the management of human diseases in Nyeri County, Kenya. CELLMED. 2016;6(3):21.1-.15.
133. Trotter RT, Logan MH. Informant consensus: a new approach for identifying potentially effective medicinal plants. Plants in Indigenous Medicine & Diet: Routledge; 2019. p. 91-112.
134. Bonet MA, Parada M, Selga A, Vallès J. Studies on pharmaceutical ethnobotany in the regions of L'Alt Empordà and Les Guilleries (Catalonia, Iberian Peninsula). J Ethnopharmacol. 1999;68(1-3):145-68.
135. Au DT, Wu J, Jiang Z, Chen H, Lu G, Zhao Z. Ethnobotanical study of medicinal plants used by Hakka in Guangdong, China. J Ethnopharmacol. 2008;117(1):41-50.
136. Girardi C, Butaud JF, Ollier C, Ingert N, Weniger B, Raharivelomanana P, et al. Herbal medicine in the Marquesas Islands. J Ethnopharmacol. 2015;161:200-13.
137. Boudjelal A, Henchiri C, Sari M, Sarri D, Hendel N, Benkhaled A, et al. Herbalists and wild medicinal plants in M'Sila (North Algeria): An ethnopharmacology survey. Journal of ethnopharmacology. 2013;148(2):395-402.
138. Malan DF, Neuba DF, Kouakou KL. Medicinal plants and traditional healing practices in Ehotile people, around the Aby Lagoon (eastern littoral of Côte d'Ivoire). J Ethnobiol Ethnomed. 2015;11:21.
139. Odongo E, Mungai N, Mutai P, Karumi E, Mwangi J, Omale J. Ethnobotanical survey of the medicinal plants used in Kakamega County, Western Kenya. Applied Medical Research. 2018;4(1):22.
140. Fadili K, Sekkate C, Alistiqsa F, Haloui Z, Chakir S, Zair T. Ethnobotanical study of medicinal plants from Er-Rich region (Moroccan high atlas). Advances in Environmental Biology. 2017;11(6):27-41.
141. Tyler SE. The work surfaces of morphogenesis: the role of the morphogenetic field. Biological Theory. 2014;9(2):194-208.
142. Nyakudya TT, Tshabalala T, Dangarembizi R, Erlwanger KH, Ndhlala AR. The Potential Therapeutic Value of Medicinal Plants in the Management of Metabolic Disorders. Molecules. 2020;25(11).
143. Mesiti F, Chavarria D, Gaspar A, Alcaro S, Borges F. The chemistry toolbox of multitarget-directed ligands for Alzheimer's disease. Eur J Med Chem. 2019;181:111572.
144. Cummings JL, Tong G, Ballard C. Treatment Combinations for Alzheimer's Disease: Current and Future Pharmacotherapy Options. J Alzheimers Dis. 2019;67(3):779-94.
145. Roy K. Multi-target drug design using chem-bioinformatic approaches: Springer; 2019.
146. Gómez-Estrada H, Díaz-Castillo F, Franco-Ospina L, Mercado-Camargo J, Guzmán-Ledezma J, Medina JD, et al. Folk medicine in the northern coast of Colombia: an overview. J Ethnobiol Ethnomed. 2011;7:27.
147. Upasani MS, Upasani SV, Beldar VG, Beldar CG, Gujarathi PP. Infrequent use of medicinal plants from India in snakebite treatment. Integr Med Res. 2018;7(1):9-26.
148. Elufioye T, Oladele A, Cyril-Olutayo C, Agbedahunsi J, Adesanya S. Ethnomedicinal study and screening of plants used for memory enhancement and antiaging in Sagamu, Nigeria. European Journal of Medicinal Plants. 2012:262-75.
149. Babawale OP, Taiye FR, Adetunji OS. Ethnobotanical survey of plants used as memory enhancer in three states of southwestern Nigeria. Journal of Applied Pharmaceutical Science. 2016;6(09):209-14.

150. Kantati YT, Kodjo KM, Dogbeavou KS, Vaudry D, Leprince J, Gbeassor M. Ethnopharmacological survey of plant species used in folk medicine against central nervous system disorders in Togo. J Ethnopharmacol. 2016;181:214-20.
151. Abu-Rabia A. Ethno-botanic treatments for paralysis (falij) in the Middle East. Chinese Medicine. 2012;3(04).
152. Mikawlrawng K, Rani R, Kumar S, Bhardwaj AR, Prakash G. Anti-paralytic medicinal plants - Review. J Tradit Complement Med. 2018;8(1):4-10.
153. Collins SW, Martins X, Mitchell A, Teshome A, Arnason JT. Fataluku medicinal ethnobotany and the East Timorese military resistance. J Ethnobiol Ethnomed. 2007;3:5.
154. Goleniowski ME, Bongiovanni GA, Palacio L, Nuñez CO, Cantero JJ. Medicinal plants from the "Sierra de Comechingones", Argentina. J Ethnopharmacol. 2006;107(3):324-41.
155. Pedrollo CT, Kinupp VF, Shepard G, Heinrich M. Medicinal plants at Rio Jauaperi, Brazilian Amazon: Ethnobotanical survey and environmental conservation. J Ethnopharmacol. 2016;186:111-24.
156. Rai PK, Lalramnghinglova H. Ethnomedicinal plant resources of Mizoram, India: Implication of traditional knowledge in health care system. Ethnobotanical Leaflets. 2010;2010(3):6.
157. Pieroni A, Sõukand R. The disappearing wild food and medicinal plant knowledge in a few mountain villages of North-Eastern Albania. Journal of Applied Botany and Food Quality. 2017;90.
158. Tounekti T, Mahdhi M, Khemira H. Ethnobotanical Study of Indigenous Medicinal Plants of Jazan Region, Saudi Arabia. Evid Based Complement Alternat Med. 2019;2019:3190670.
159. Bhattarai S, Chaudhary RP, Taylor RS. Ethnomedicinal plants used by the people of Manang district, central Nepal. J Ethnobiol Ethnomed. 2006;2:41.
160. Sharma J, Gairola S, Gaur RD, Painuli RM, Siddiqi TO. Ethnomedicinal plants used for treating epilepsy by indigenous communities of sub-Himalayan region of Uttarakhand, India. J Ethnopharmacol. 2013;150(1):353-70.
161. Getaneh S, Girma Z. An ethnobotanical study of medicinal plants in Debre Libanos Wereda, Central Ethiopia. African Journal of Plant Science. 2014;8(7):366-79.
162. Longuefosse JL, Nossin E. Medical ethnobotany survey in Martinique. J Ethnopharmacol. 1996;53(3):117-42.
163. Malawani A, Nuneza O, Uy M, Senarath W. Ethnobotanical survey of the medicinal plants used by the Maranois in Pualas, Lanao del Sur. Philippines BEPLS. 2017;6(6):45-53.
164. Motti R, Motti P. An ethnobotanical survey of useful plants in the agro Nocerino Sarnese (Campania, southern Italy). Human Ecology. 2017;45(6):865-78.
165. Gazzaneo LRS, De Lucena RFP, de Albuquerque UP. Knowledge and use of medicinal plants by local specialists in a region of Atlantic Forest in the state of Pernambuco (Northeastern Brazil). Journal of ethnobiology and ethnomedicine. 2005;1(1):1-8.
166. Yineger H, Yewhalaw D. Traditional medicinal plant knowledge and use by local healers in Sekoru District, Jimma Zone, Southwestern Ethiopia. J Ethnobiol Ethnomed. 2007;3:24.
167. Bekalo TH, Woodmatas SD, Woldemariam ZA. An ethnobotanical study of medicinal plants used by local people in the lowlands of Konta Special Woreda, southern nations, nationalities and peoples regional state, Ethiopia. Journal of Ethnobiology and Ethnomedicine. 2009;5(1):1-15.
168. Lulekal E, Kelbessa E, Bekele T, Yineger H. An ethnobotanical study of medicinal plants in Mana Angetu district, southeastern Ethiopia. J Ethnobiol Ethnomed. 2008;4:10.
169. Polat R, Satıl F. An ethnobotanical survey of medicinal plants in Edremit Gulf (Balıkesir-Turkey). J Ethnopharmacol. 2012;139(2):626-41.
170. Bussmann RW, Sharon D. Traditional medicinal plant use in Northern Peru: tracking two thousand years of healing culture. J Ethnobiol Ethnomed. 2006;2:47.
171. Lee C, Kim S-Y, Eum S, Paik J-H, Bach TT, Darshetkar AM, et al. Ethnobotanical study on medicinal plants used by local Van Kieu ethnic people of Bac Huong Hoa nature reserve, Vietnam. Journal of ethnopharmacology. 2019;231:283-94.
172. Ji H, Shengji P, Chunlin L. An ethnobotanical study of medicinal plants used by the Lisu people in Nujiang, northwest Yunnan, China. Economic botany. 2004;58(1):S253-S64.
173. Bourdy G, DeWalt SJ, Chávez de Michel LR, Roca A, Deharo E, Muñoz V, et al. Medicinal plants uses of the Tacana, an Amazonian Bolivian ethnic group. J Ethnopharmacol. 2000;70(2):87-109.

174. Sun W, Yan B, Wang R, Liu F, Hu Z, Zhou L, et al. In vivo acute toxicity of detoxified Fuzi (lateral root of *Aconitum carmichaeli*) after a traditional detoxification process. EXCLI J. 2018;17:889-99.
175. Jayasinghe CD, Jayawardena UA. Toxicity Assessment of Herbal Medicine Using Zebrafish Embryos: A Systematic Review. Evid Based Complement Alternat Med. 2019;2019:7272808.
176. Alafiatayo AA, Lai K-S, Syahida A, Mahmood M, Shaharuddin NA. Phytochemical evaluation, embryotoxicity, and teratogenic effects of Curcuma longa extract on zebrafish (Danio rerio). Evidence-Based Complementary and Alternative Medicine. 2019;2019.
177. Voeks RA, Leony A. Forgetting the forest: assessing medicinal plant erosion in eastern Brazil. Economic Botany. 2004;58(1):S294-S306.
178. Nankaya J, Gichuki N, Lukhoba C, Balslev H. Sustainability of the loita Maasai childrens' ethnomedicinal knowledge. Sustainability. 2019;11(19):5530.
179. Begossi A, Hanazaki N, Tamashiro JY. Medicinal plants in the Atlantic Forest (Brazil): knowledge, use, and conservation. Human ecology. 2002;30(3):281-99.
180. Joly LG, Guerra S, Séptimo R, Solís PN, Correa M, Gupta M, et al. Ethnobotanical inventory of medicinal plants used by the Guaymi Indians in western Panama. Part I. J Ethnopharmacol. 1987;20(2):145-71.
181. Chander MP, Kartick C, Gangadhar J, Vijayachari P. Ethno medicine and healthcare practices among Nicobarese of Car Nicobar - an indigenous tribe of Andaman and Nicobar Islands. J Ethnopharmacol. 2014;158 Pt A:18-24.
182. Kassam K-A. Viewing change through the prism of indigenous human ecology: findings from the Afghan and Tajik Pamirs. Human Ecology. 2009;37(6):677-90.
183. Pawera L, Verner V, Termote C, Sodombekov I, Kandakov A, Karabaev N, et al. Medical ethnobotany of herbal practitioners in the Turkestan Range, southwestern Kyrgyzstan. Acta Societatis Botanicorum Poloniae. 2016;85(1).