Wilson disease

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Abstract | Wilson disease (WD) is a potentially treatable, inherited disorder of copper metabolism that is characterized by the pathological accumulation of copper. WD is caused by mutations in *ATP7B*, which encodes a transmembrane copper-transporting ATPase, leading to impaired copper homeostasis and copper overload in the liver, brain and other organs. The clinical course of WD can vary in the type and severity of symptoms, but progressive liver disease is a common feature. Patients can also present with neurological disorders and psychiatric symptoms. WD is diagnosed using diagnostic algorithms that incorporate clinical symptoms and signs, measures of copper metabolism and DNA analysis of *ATP7B*. Available treatments include chelation therapy and zinc salts, which reverse copper overload by different mechanisms. Additionally, liver transplantation is indicated in selected cases. New agents, such as tetrathiomolybdate salts, are currently being investigated in clinical trials, and genetic therapies are being tested in animal models. With early diagnosis and treatment, the prognosis is good; however, an important issue is diagnosing patients before the onset of serious symptoms. Advances in screening for WD may therefore bring earlier diagnosis and improvements for patients with WD.

Wilson disease (WD) is an inherited disorder of copper metabolism[1]. The disease is caused by homozygous or compound heterozygous mutations (the presence of two different mutant alleles) in *ATP7B*, which encodes transmembrane copper-transporting ATPase 2 (widely known as ATP7B), which mediates the excretion of copper into bile and delivers copper for the functional synthesis of ceruloplasmin (the major copper-transporting protein in the blood)[2]. The liver is the site of metabolism for dietary copper; in WD, defective ATP7B function leads to copper overload in hepatocytes, which is associated with liver pathology. Excess non-ceruloplasmin-bound copper is also released into the circulation, with secondary pathological accumulation in other tissues, particularly the brain, which can lead to neurological symptoms and psychiatric disturbances. Symptoms vary widely and present most commonly between 5 and 35 years of age. WD is rare, with the prevalence of symptomatic disease estimated to be 1 case per ~30,000 individuals; however, a greater prevalence of genetic WD (based on two alleles with pathogenetic mutations) has been observed according to recent molecular studies. WD belongs in a category with only a few other genetic disorders that can be successfully managed if diagnosed early and correctly treated[1,3]; however, if untreated, WD is universally fatal[4–6]. A timeline of important discoveries in WD is shown in FIG. 1.

In this Primer, we summarize current knowledge on WD, covering epidemiology, genetics, pathogenesis, clinical manifestations and diagnosis, and discuss existing management options as well as future treatment possibilities.

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Epidemiology

In the 1970s, epidemiological studies indicated a WD prevalence of 29 cases per 1,000,000 individuals in Germany and 33 cases per 1,000,000 individuals in Japan[7,8]. In 1984, the global prevalence of WD was estimated as 1 case per 30,000 individuals in non-isolated populations, with a mutation carrier frequency (individuals with one disease-associated allele) of 1 in 90 individuals[9] (which corresponds to almost 1% of the general population); these epidemiological data are still widely cited today. The prevalence of WD is higher in China (58.7 cases per 1,000,000 individuals) and Asian countries than in Western countries[10]. Epidemiological studies from isolated communities reported a higher frequency of WD due to consanguinity (for example, the Canary Islands report 1 case per 2,600 individuals; Sardinia reports 1 case per 7,000 individuals)[4,11,12]. Furthermore, in a molecular study from the UK, the calculated frequency of individuals predicted to carry two mutant pathogenetic *ATP7B* alleles was ~1 per 7,000 individuals, with heterozygote mutations potentially being found in up to 2.5% of the general population[12]. An underestimation of the prevalence of WD may be associated with the varying clinical presentation of disease, leading to underdiagnosis and misdiagnosis, the low sensitivity of certain copper metabolism tests and the unknown age-related clinical penetrance of *ATP7B*

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mutations. With the increasing awareness of WD among physicians and the increasing availability of genetic tests, the number of patients diagnosed with WD appears to be rising. New genetic methods, including entire *ATP7B* gene sequencing, are expensive and not readily available in all countries but are useful when available to further define prevalence.

Mortality data in presymptomatic patients with WD who are treatment adherent are comparable to the general population[3]. However, in the entire population of patients with WD (regardless of adherence, clinical symptoms, advancement of disease at diagnosis or type of treatment), studies commonly show that mortality in patients with WD is 5–6.1% higher than in the general population[13–15]. The presence of advanced hepatic and neurological disease, and the lack of adherence of patients to treatment affect survival.

Mechanisms/pathophysiology

In WD, mutations in *ATP7B* and inactivation of the ATP7B transporter in hepatocytes result in the failure of the biliary excretion of copper, which leads to disturbed copper homeostasis. ATP7B is also responsible for transporting copper for the synthesis of functional ceruloplasmin. In the blood, functional ceruloplasmin contains six copper atoms per molecule (holoceruloplasmin), but the protein may be present without bound copper (apoceruloplasmin). In patients with WD, impaired ATP7B results in decreased serum ceruloplasmin levels, and levels of total serum copper may be decreased compared with healthy controls; however, levels of toxic non-ceruloplasmin-bound copper are often increased[4]. Hepatic and systemic overload of toxic copper is the major cause of tissue pathology and clinical symptoms in patients.

Genetics

Mutations in ATP7B. WD is an autosomal recessive disease caused by mutations in *ATP7B* (BOX 1), which is located on the short arm of chromosome 13 and contains 20 introns and 21 exons. More than 700 mutations have been described in *ATP7B* according to the Human Gene Mutation Database[16,17]; patients with WD can be homozygous for one disease-causing mutation or carry two different disease-causing mutations as compound heterozygotes. WD-associated mutations can affect almost all 21 exons and are frequently missense and nonsense. The missense mutation H1069Q in exon 14 is very common; ~50–80% of patients with WD from central, eastern and northern Europe carry at least one allele with the H1069Q mutation[2]. In southern Europe, other mutations are common, such as the missense mutation M645R in mainland Spain[2]. By contrast, in southeastern Asia, the R778L mutation in exon 8 is found more frequently and has an allele frequency of 14–49% in patients with WD[2].

Several studies have attempted the challenging task of correlating *ATP7B* genotype with WD phenotype. In vitro experiments demonstrated that different *ATP7B* variants present different functional properties with varying copper transporter activity[18]. Studies in the 2000s suggested that homozygosity of the H1069Q mutation leads to later onset of WD and more frequent neurological presentations than H1069Q compound heterozygotes[19–21] and that patients with WD with frameshift and nonsense mutations in *ATP7B* had lower serum ceruloplasmin levels than those with missense mutations[22]. However, the latter observation was not confirmed by larger studies[23]. In a small sample of patients with WD, it was suggested that certain mutations (named 'truncated mutations') are associated with patients presenting with acute liver failure and an earlier age of disease onset. However, overall, results from studies attempting genotype–phenotype correlations have not been conclusive[23–28], partly owing to the poor phenotypic characterization of patients with WD, late diagnosis and overlapping neurological, psychiatric and hepatic signs and symptoms of various severities. Genetic and environmental factors likely interact to influence the complex disease phenotype; however, further studies are required to provide conclusive evidence for specific associations.

Other genes. Studies have explored the role of proteins and mutated genes other than *ATP7B* as contributors to the phenotype of WD. Patatin-like phospholipase domain-containing protein 3 (PNPLA3) is involved in triglyceride metabolism; a *PNPLA3* variant most commonly associated with non-alcoholic fatty liver disease (NAFLD) has also been linked with severity of hepatic steatosis (fat accumulation in the liver) in patients with WD[29]. A relationship between PNPLA3 loss of function and accumulation of triglycerides in hepatocytes and hepatic stellate cells has also been observed[30]. A second gene involved in lipid metabolism, the ε4 allele of the apolipoprotein E gene (*APOE*), which is associated with neurodegenerative diseases, was proposed to be a modifier of the phenotype of WD[31], but a large study showed that the *APOE* ε4 genotype had no association with either the hepatic or neurological phenotype in WD[32]. However, *APOE*-ε4-positive women tended to have disease onset at a younger age than women with the ε3/ε3 genotype, particularly when they were also homozygous for the H1069Q mutation, which suggests a faster progression of disease in these patients[32]. Interestingly, mutations in the copper metabolism domain containing 1 (*COMMD1*) gene (formerly *MURR1*) are the cause of copper accumulation in Bedlington terriers,

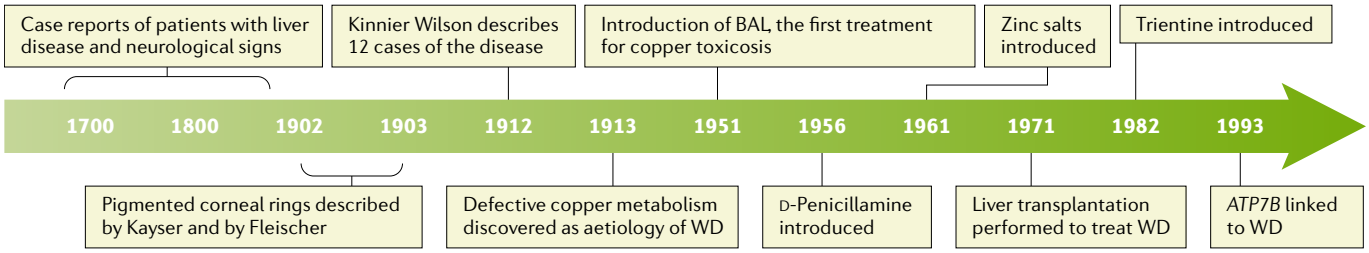


Fig. 1 | **A timeline of key discoveries in WD.** Samuel Alexander Kinnier Wilson described Wilson disease (WD) in 12 patients in 1912 in terms of a heredity condition involving the co-occurrence of liver cirrhosis and neurological deficits. However, the first cases of WD with dominant tremor symptoms were described in 1883 by Carl Westphal, whereas the corneal rings that are characteristic of WD were described by Bernhard Kayser and by Bruno Fleischer in 1902–1903. Subsequently, disturbances in copper metabolism were discovered as the aetiology of WD, and it was identified as an autosomal recessive inherited disorder. In 1951, the chelator British anti-Lewisite (BAL) became the first treatment for copper toxicosis and for the treatment of WD. In 1956, the chelator, D-penicillamine, introduced by John Walshe, became the first oral drug for WD, which was followed by the availability of zinc salts in 1961 and trientine (another chelator) in 1982. Liver transplantation, as an ultimate treatment for WD, was performed by Thomas Starzl and colleagues in 1971. In 1993, the gene associated with WD, *ATP7B*, was located to chromosome 13q and found to code a P-type ATPase involved in copper transport[209].

and one study identified *COMMD1* variants in 30% of a small cohort of 63 patients with WD[33]. However, later studies on different patient populations could not confirm these findings[34,35]. The copper transport protein ATOX1 has also garnered much attention given its interaction with ATP7B[36], but correlational studies on patients with WD could not identify a significant role for *ATOX1* mutations[37–39].

Oxidative stress caused by reactive oxygen species is thought to be the main cause of liver damage associated with copper accumulation. In a cohort of 435 patients with WD, variants of genes that encode antioxidant enzymes, including catalase and manganese superoxide dismutase, were linked to the age of onset of WD[40]. Variants of the gene encoding methylenetetrahydrofolate reductase (*MTHFR*), a key folate–homocysteine pathway enzyme, were studied in a large group of Polish patients with WD. Although the study was criticized for lack of phenotypic detail, the authors identified a correlation between WD phenotype and *MTHFR* polymorphisms[41]; patients with the *MTHFR* 677T allele more frequently exhibited liver disease. Despite the limitations of the study, *MTHFR* represents an interesting gene for further study as mutations can influence folate and methionine metabolism, which has possible downstream effects on epigenetic mechanisms of gene expression and regulation.

Epigenetics

Indirect data from patients with WD and studies in animal models indicate that epigenetic mechanisms may be involved in the pathogenesis of WD and its phenotypic presentation. Interestingly, several case reports describe homozygous twins with WD who present with different disease phenotypes[42–44], suggesting that environmental or nutritional factors may affect WD. The potential role of such factors on epigenetic mechanisms has been explored in animal models of WD. At the interface between the regulation of gene expression and the environment is methionine metabolism, a metabolic pathway that has regulatory effects on DNA methylation. The enzyme *S*-adenosylhomocysteine (SAH) hydrolase (SAHH; also known as AHCY) has a crucial role in methionine metabolism as it is responsible for metabolizing SAH to homocysteine. If the expression or activity of SAHH are decreased, the level of SAH, which acts as an inhibitor of DNA methylation reactions, will increase. Importantly, SAHH enzyme activity and gene (*AHCY*) transcript levels are decreased in the presence of hepatic copper accumulation with consequent downstream changes in methionine metabolism parameters[45,46]. Notably, the toxic-milk mouse from the Jackson Laboratory (tx-j mouse), which has a spontaneous point mutation affecting the second transmembrane region of the copper transporter, showed dysregulation of methionine metabolism and global DNA hypomethylation in hepatocytes[47], with possible downstream effects on the regulation of genes involved in the development of liver damage. In addition, during embryonic development, the liver (a site of major methylation rearrangements) presented major changes in gene transcript levels related to cell cycle and replication in tx-j mice compared with control animals[48]. The provision of supplemental methyl donor choline to pregnant mice was able to bring gene expression in embryonic mice to the same levels as control animals, indicating that fetal livers are susceptible to nutritional factors with potential lifelong consequences on disease phenotype and progression[48].

Copper homeostasis

Copper is essential for human physiology: copper serves as a cofactor for various enzymes, such as those that are critically involved in respiration (for example, cytochrome *c* oxidase), activation of neuroendocrine peptides (for example, peptidyl-α-monooxygenase), pigmentation (for example, tyrosinase), catecholamine synthesis and clearance (for example, dopamine-β-monooxygenase), free radical defence (for example, superoxide dismutase 1 (SOD1) and SOD3) and enzymes involved in many other cellular processes. Normal dietary consumption and absorption of copper, which is contributed mainly by legumes, potatoes, nuts and seeds, chocolate, beef, organ

meat and shellfish[49], exceed the metabolic demand, therefore appropriate levels are controlled by regulation of the biliary excretion of copper[4,50].

Copper homeostasis is maintained by a network of proteins, which includes transmembrane copper transporters, cytosolic copper carrier proteins, copper storage molecules (metallothioneins) and copper-requiring enzymes. In addition, proteins that do not bind copper directly, but regulate the abundance or activity of the copper-binding and/or transporting proteins, also contribute to cellular copper homeostasis. This regulatory network includes adaptor proteins, kinases, components of the cellular trafficking machinery and DNA- and RNA-binding proteins. The mechanisms regulating copper homeostasis are cell-type-specific; therefore, the abundance, distribution and cellular behaviour of the major copper homeostatic molecules and their regulators differ between cell types. Nevertheless, the same core protein framework regulates copper homeostasis in most cells.

Copper primarily enters cells through the high affinity copper uptake protein 1 (CTR1; also known as SLC31A1)[51–53] (FIG. 2). Copper chaperones shuttle copper to specific intracellular targets; for example, copper chaperone for SOD (CCS) transports copper to SOD1 whereas ATOX1 shuttles copper to the copper-transporting ATPase 1 (ATP7A) and ATP7B transporters. ATP7A and ATP7B transport copper into the trans-Golgi network for subsequent incorporation into copper-dependent enzymes and to the cellular membrane for the excretion of excessive copper.

Hepatocytes are the site of two important physiological processes in copper homeostasis: first, ATP7B provides copper for incorporation into apoceruloplasmin for the synthesis of functional (holo)ceruloplasmin; and second, ATP7B facilitates the process of biliary copper excretion (FIG. 2). Inactivation of ATP7A or ATP7B results in marked disturbances in copper homeostasis and the inactivation of specific copper-dependent enzymes and manifests clinically as Menkes disease (BOX 2) or WD, respectively. In addition, MEDNIK (mental retardation, enteropathy, deafness, neuropathy, ichthyosis and keratoderma) syndrome is caused by mutations in the gene for an adaptor protein that participates in the intracellular trafficking of ATP7A and ATP7B (BOX 2).

Mitochondria use cellular copper for respiration and are key regulators of the cellular copper balance[54]. How copper is distributed between the cytosolic copper proteins and the copper-binding proteins in mitochondria is unclear. The current model suggests that a gradient of protein–copper binding affinities and, presumably, the relative abundance of copper-binding proteins governs the partitioning of copper between cytosolic proteins and mitochondrial proteins[55]. Inhibitory mutations in SCO1 and SCO2 (protein SCO1 and SCO2, mitochondrial, respectively), which facilitate copper incorporation into cytochrome *c* oxidase, result in mitochondrial dysfunction and altered cellular copper homeostasis[54]. As knowledge of the overall copper homeostatic network continues to expand, the link to numerous cellular processes becomes more and more apparent.

Recently, new and intriguing roles for copper have emerged in normal physiological processes and the pathophysiology of disease. For example, it became apparent that copper misbalance is a contributing factor to lipid dyshomeostasis[56,57]. Abnormal lipid metabolism associated with either copper overload or deficiency is commonly observed in disorders such as WD, NAFLD and diabetes mellitus[56,58–60]. In addition, important physiological processes such as the assembly of chylomicrons (triglyceride-rich lipoproteins), blood vessel formation, myelination of neurons, wound healing and the immune response depend on copper homeostasis[61–64]. Moreover, the role of copper in cell proliferation and angiogenesis is finding its first applications in clinical practice as the copper-protein-binding agent, tetrathiomolybdate, is being evaluated in patients with cancer[65].

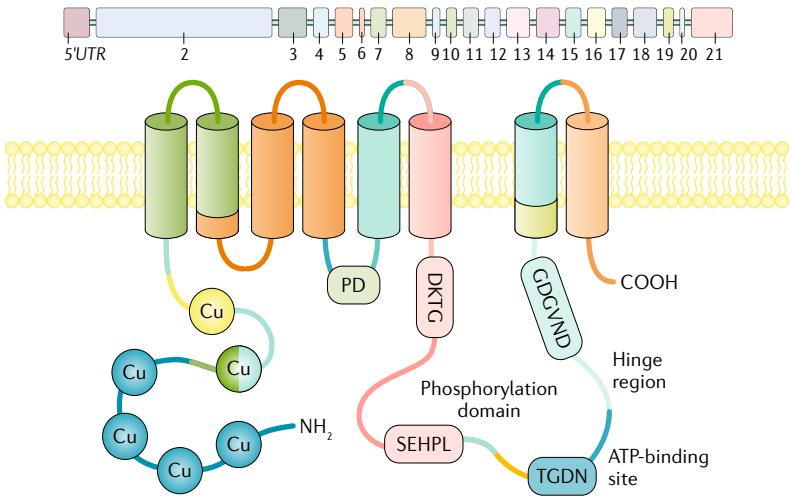
Pathogenesis

The primary cause of clinical symptoms in WD are pathological tissue changes triggered by the toxic effects of excess copper[66] (FIG. 3). In WD, labile non-ceruloplasmin-bound copper (that is, the pool of copper ions loosely bound to serum albumin and other molecules that can easily engage

Box 1 | ATP7B structure

Transmembrane copper-transporting ATPase 2 (ATP7B) belongs to class 1B of the highly conserved P-type ATPase superfamily responsible for the transport of copper and other heavy metals across cellular membranes. The protein contains 1,465 amino acids organized into the phosphatase domain (PD), the phosphorylation domain and eight transmembrane ion channels that span the phospholipid bilayer of the plasma membrane (see the figure; colour-coding in the figure represents different exons in *ATP7B*). The copper (Cu)-binding domain is composed of six copper-binding sites, which play a central role in accepting Cu from the copper transport protein ATOX1 through protein–protein interactions[207].

Different *ATP7B* mutations affect the function of ATP7B in different ways. One of the most common mutations in Wilson disease, H1069Q, occurs in the SEHPL motif, resulting in protein misfolding, abnormal phosphorylation in the phosphorylation domain, decreased ATP-binding affinity, thermal instability and abnormal localization to the trans-Golgi network[207]. The E1064A mutation, also found in the SEHPL motif, completely disables ATP-binding affinity but does not result in protein misfolding, transport abnormalities or thermal instability[207]. The R778L mutation affects the transmembrane transport of copper whereas the G943S and M769V mutations result in defective copper metabolism but preserved ceruloplasmin levels[207]. Other prevalent mutations, such as protein-truncating nonsense mutations and frameshift mutations, are predicted to cause decay of *ATP7B* mRNA or a severely truncated protein, resulting in absent or diminished levels of functional ATP7B[207].



COOH, carboxyl terminus; NH_2, amino terminus. Figure adapted with permission from REF.[207], Elsevier.

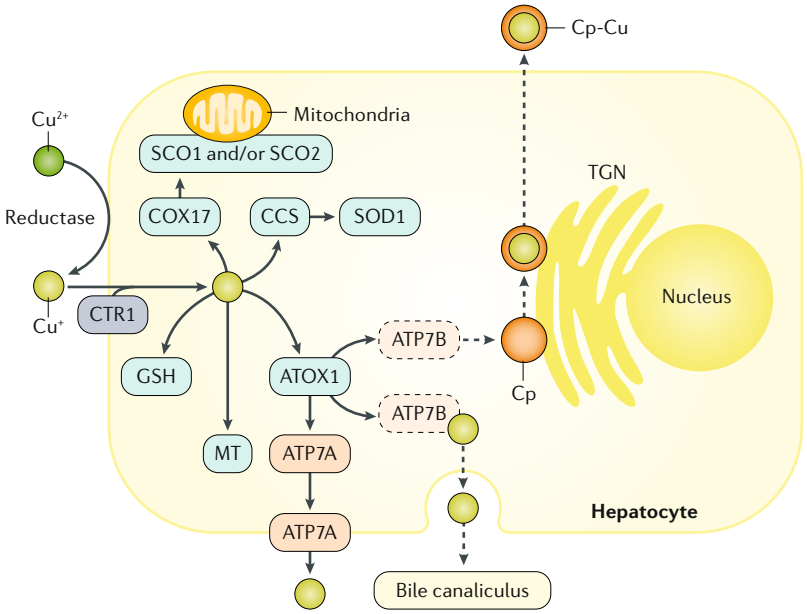


Fig. 2 | **Copper homeostasis in hepatocytes.** Cellular copper (Cu) uptake in hepatocytes and other cells is primarily mediated by high affinity copper uptake protein 1 (CTR1). A yet-unknown cuprireductase and/or extracellular ascorbate provide the reduced Cu species for cellular uptake by CTR1. Specialized chaperones shuttle Cu to its specific intracellular targets: the Cu chaperone for superoxide dismutase (CCS) shuttles Cu to superoxide dismutase 1 (SOD1); cytochrome *c* oxidase copper chaperone (COX17) shuttles Cu to protein SCO1 and/or SCO2, mitochondrial (SCO1 and SCO2, respectively), for subsequent incorporation into cytochrome *c* oxidase; and copper transport protein ATOX1 (ATOX1) shuttles Cu to the Cu-transporting ATPases, copper-transporting ATPase 1 (ATP7A) and transmembrane copper-transporting ATPase 2 (ATP7B), in the trans-Golgi network (TGN). ATP7B has two functions in hepatocytes: first, in the TGN, ATP7B activates ceruloplasmin (Cp) by packaging six Cu molecules into apoceruloplasmin, which is then secreted into the plasma; second, in the cytoplasm, ATP7B sequesters excess Cu into vesicles and excretes it across the apical membrane into bile[77]. Thus, in the liver, ATP7B provides Cu for incorporation into Cp and is also required for biliary Cu excretion. The pathways altered in Wilson disease (WD) are marked by dashed lines. With reduced or absent levels of ATP7B in WD, there is reduced biliary Cu excretion and reduced incorporation of Cu into Cp[77]. GSH, glutathione; MT, metallothioneins.

in chemical reactions) present in blood is continuously taken up by virtually all tissues, possibly via CTR1 and divalent metal transporter 1 (DMT1; also known as natural resistance-associated macrophage protein 2 or SLC11A2). Evidence suggests that DMT1 transports copper ions intracellularly even when copper is in excess[67].

The toxicity of excess copper in tissues is likely a consequence of its redox activity, which leads to oxidative stress and the subsequent damage of lipids, proteins, DNA and RNA molecules. Other possible mechanisms of copper toxicity include the induction of apoptosis by activation of acid sphingomyelinase, which triggers the release of the apoptotic secondary messenger ceramide[68], as well as direct inhibition of enzymatic activities through its nonspecific binding to protein thiol groups[69,70]. At the subcellular level, mitochondria are the most sensitive targets for copper-induced toxicity[71,72].

Liver. The liver has the highest tissue expression of the ATP7B copper transporter and is the central organ that regulates systemic copper homeostasis. Impairment of copper excretion caused by ATP7B dysfunction leads to hepatic copper accumulation. Liver injury is therefore the earliest and most frequent manifestation of WD. Hepatic copper concentration in patients with WD is typically increased around fivefold compared with that in healthy individuals[4]. Copper is not distributed homogeneously within the liver, and its cellular localization also varies during disease progression. In the initial stages of WD, copper is present diffusely in the cytoplasm of hepatocytes bound to metallothioneins, which are cysteine-rich proteins with the ability to bind, store and detoxify heavy metals. As copper accumulates in hepatocyte lysosomes, it may become detectable by stains such as Timm stain, rhodanine (FIG. 4) and orcein[73]. Adverse effects on the integrity and function of hepatocyte mitochondria can be detected early in the disease course[74]. Mitochondrial damage can result in impaired hepatic energy metabolism and the downregulation of genes involved in cholesterol biosynthesis, which both contribute to hepatic steatosis (FIG. 4). Chronic hepatocyte injury and cell death ultimately lead to inflammatory changes (hepatitis) and the net accumulation of extracellular matrix (fibrosis) within the liver. Notably, apoptosis is an important cause of hepatocyte loss and may be triggered by cytochrome *c* released from damaged mitochondria or through the activation of acid sphingomyelinase by copper and the release of ceramide[68,74].

In WD, several types of liver pathology may be observed by light microscopy, including glycogenated hepatocytic nuclei (that is, typically enlarged nuclei with optically clear intranuclear inclusions with accentuated nuclear membrane, which are caused by the high glycogen content), Mallory-Denk bodies (cellular inclusions composed of misfolded cytoskeletal elements including keratin and ubiquitin-binding protein p62) and portal and lobular inflammation in combination with focal or diffuse hepatocyte steatosis[75]. In general, microscopic findings in WD are not specific; at the initial stage of liver affliction, liver pathology may strikingly resemble NAFLD. With progressive damage, hepatic fibrosis and subsequently macronodular cirrhosis typically develop.

If WD is untreated, the disease progresses and the copper storage capacity of hepatocytes becomes exhausted; therefore, ingested and absorbed copper cannot be further sequestered by the liver. At this point, the amount of labile non-ceruloplasmin-bound copper increases in the bloodstream[76]. Gradually, copper absorbed from the diet and copper released from hepatocytes with exhausted endogenous copper storage capacity progressively accumulate in other organs, most notably in the brain, eyes, kidneys, bones and heart, exerting extrahepatic toxicity. In addition, rapid copper release from the liver can occur owing to mass necrosis of hepatocytes[77].

Brain. The concentration of copper in the brain of patients with WD may reach values 10–15 times higher than in control individuals[78,79]. The connection between copper deposits and cerebral tissue damage was confirmed in a study of the post-mortem brains of 11 patients with WD, which showed a fair degree of correlation between cerebral copper content and the severity of neuropathology[79]. In the brain, the toxic effect of copper is first buffered by astrocytes; upon taking up

Box 2 | **Other genetic disorders of copper metabolism**

The X-linked recessive disorder, Menkes disease, is characterized by impaired copper absorption due to mutations in *ATP7A*[1], which encodes for copper-transporting ATPase 1 (widely known as ATP7A). Menkes disease typically presents in boys aged 2–3 months and is characterized by neurodevelopmental delay and degeneration, seizures and failure to thrive. Without early recognition and treatment with parenteral copper replacement, death usually occurs several years after onset. Occipital horn syndrome is caused by another allelic variant of *ATP7A*, with a less-severe neurological phenotype (slight generalized muscle weakness, dysautonomia (including syncope), orthostatic hypotension and chronic diarrhoea) that is often not diagnosed until mid-childhood or later. Another *ATP7A* allelic variant, distal motor neuropathy without overt copper metabolic abnormalities, presents with progressive distal motor neuropathy (curled fingers, foot deformities and diminished deep tendon reflexes), with minimal or no sensory symptoms.

Huppke-Brendel syndrome results from mutations in *SLC33A1*, which encodes the acetyl-coenzyme A transporter 1. This syndrome is characterized by low serum copper and ceruloplasmin concentrations, alongside congenital cataracts, hearing loss and severe developmental delay. Similar to Menkes disease, pronounced cerebellar hypoplasia and hypomyelination are also seen.

The newly recognized copper metabolism disorder, MEDNIK (mental retardation, enteropathy, deafness, neuropathy, ichthyosis and keratoderma) syndrome, is an autosomal recessive trait caused by mutations in the *AP1S1* gene, which encodes the σ1A small subunit of the adaptor protein complex 1 (AP1) that is involved in intracellular trafficking of transmembrane proteins. Abnormalities in copper ATPase trafficking result in decreased total serum copper, decreased serum ceruloplasmin and hepatic copper accumulation, similar to that seen in WD.

Ceruloplasmin is also involved in iron metabolism, and the genetic defect aceruloplasminaemia (which is characterized by a lack of serum ceruloplasmin) presents with neurological disorders, diabetes mellitus and microcytic anaemia associated with excessive systemic iron accumulation, which is typically accompanied by low serum copper concentrations[1]. Defects in copper metabolism are also seen in some patients with congenital disorders of glycosylation and may be accompanied by severe liver disease[208].

excess copper, they increase in numbers (astrogliosis), undergo cellular swelling and upregulate synthesis of metallothionein to increase their storage capacity for copper[80,81]. Long-term exposure to high copper concentrations ultimately results in morphological changes and functional impairment of astrocytes. Astrocytes form an important part of the blood–brain barrier and are essential for neuronal homeostasis. Increased tissue copper levels and alteration of the cerebral microenvironment caused by astrocyte damage lead to affliction of other cells and tissues of the brain, including neurons and oligodendrocytes[82].

Evidence suggests that, for unknown reasons, different brain regions have distinct susceptibility to copper toxicity[83]. Pathological changes — which include astrogliosis, demyelination and tissue disintegration (ranging from mild rarefaction to necrosis) — are most often reported in the basal ganglia, thalamus, cerebellum and upper brainstem (FIG. 5); these abnormalities are depicted as T_2 hyperintense lesions on MRI. Demyelination particularly affects bundles passing through basal ganglia and pontine fibres[83,84]. In the basal ganglia, inflammatory changes (with the accumulation of heavy iron-laden macrophages) are frequently present[83]. In a neuropathological study assessing iron concentration and the degree of tissue disruption, more severe pathology in the putamen was associated with an increased number of iron-positive phagocytic and astrocytic cells[83]. Whether iron deposits causally contribute to neuropathology in WD is currently unknown[85]. As shown in FIG. 5, T_2 hypointense lesions on MRI (FIG. 5a,b) are associated with increased iron deposits (FIG. 5c,f,g).

The putamen is the most frequently and severely affected brain region in WD, with lesions linked mainly to dystonia and parkinsonism[86]. Dysfunction of the cortico-striatal pathways may lead to psychiatric symptoms and cognitive deficits mostly affecting executive function[87]. The dorsal midbrain, particularly the dentate-rubro-thalamic pathway, is another frequently affected structure, and lesions here may be associated with coarse-action tremor (kinetic or intention tremor with large displacement)[88]. Lesions in the cortex and subcortical white matter are reported only sparsely in treated patients; these lesions may be associated with epileptic seizures[89].

In addition to lesions caused by copper toxicity, hepatic encephalopathy may contribute to neuropsychiatric symptoms in WD where brain dysfunction is caused by liver insufficiency and/or portosystemic shunting (where the portal circulation with gut-derived toxins bypasses the liver and flows into the systemic circulation). This model is supported by the resemblance of neuropathological abnormalities, such as the presence of abnormal astrocytes (referred to as Alzheimer-type glia) and specific MRI findings (that is, bilateral pallidal T_1 hyperintensities) in WD and hepatic encephalopathy[77]. Morphological and functional retinal abnormalities are observed in WD and are associated with the severity of brain pathology detected by MRI and with neurological impairment[90,91].

Other organs. The pathophysiology of WD in other organs has been less well investigated. The rapid release of copper caused by mass hepatocyte necrosis in WD can lead to a considerable increase of copper blood levels within only days and therefore may mimic acute copper poisoning. It manifests as Coombs-negative (non-autoimmune) haemolytic anaemia variably accompanied by rhabdomyolysis (the breakdown of skeletal muscle tissue) and renal tubular damage. The mechanisms of haemolysis and rhabdomyolysis are not entirely understood. In erythrocytes, copper may theoretically react with membrane lipids and inhibit sulfhydryl groups of the enzymes glucose-6-phosphate dehydrogenase and glutathione reductase; these processes can reduce cellular antioxidant capacity, which may ultimately lead to oxidative damage of haemoglobin and the cell membrane[92,93]. Acute rhabdomyolysis may result from copper-induced inhibition of Na^+/K^+-ATPase activity in muscle fibres[94]. Leukopenia and thrombocytopenia are frequent findings in patients with WD, which may be ascribed to splenic sequestration of leukocytes and platelets in patients with cirrhosis-associated portal hypertension (a complication of cirrhosis in which there is an increase in the blood pressure of the portal venous system)[95,96].

Non-ceruloplasmin-bound copper in the serum is filtrated by the renal tubular epithelium and excreted via urine. However, in WD, excess copper in renal parenchyma may cause renal tubular dysfunction[97]. Furthermore, in WD, pathological changes to bone

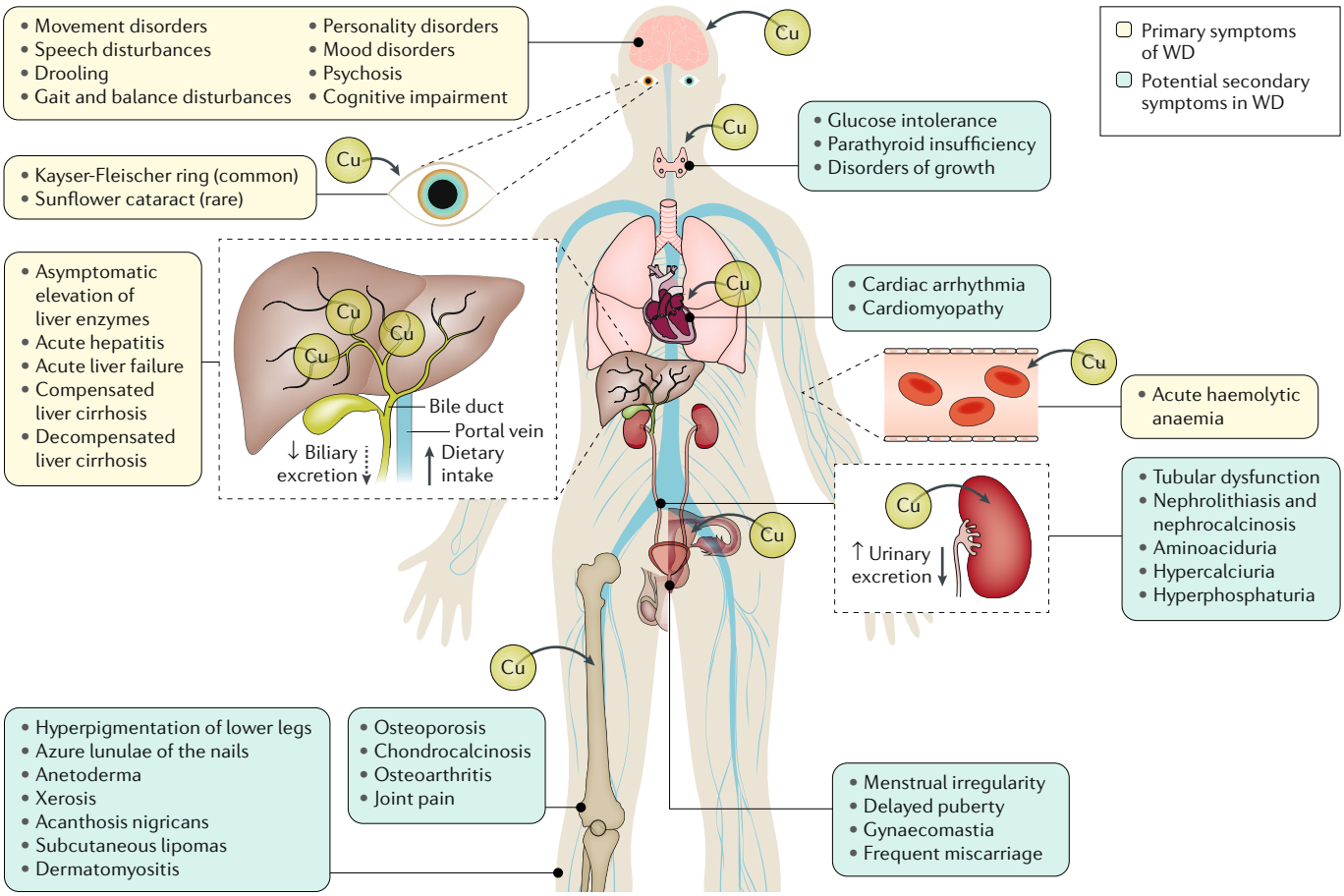


Fig. 3 | **Copper toxicity in the pathogenesis of WD.** Dietary copper (Cu) is transported via the portal vein and sequestered in the liver, which is the central organ for systemic Cu homeostasis. Impairment of biliary Cu excretion in Wilson disease (WD) leads to gradual Cu accumulation in liver. When the capacity of the liver to store Cu is exhausted, excessive quantities of non-ceruloplasmin-bound Cu enter the systemic circulation and are deposited and accumulate in various organs, exerting extrahepatic Cu toxicity. Cu accumulates in the cornea, brain, red blood cells, skeletal and cardiac muscle cells, synovial membranes of large joints and renal parenchyma, causing the various clinical manifestations of WD. The primary clinical manifestations are associated with Cu accumulation in the liver, brain and eyes. Non-ceruloplasmin-bound plasma Cu is filtrated by the renal tubular epithelium and is excreted via urine.

structure, such as osteomalacia and osteoporosis, with increased incidence of spontaneous fractures, have been observed[98]. Copper accumulation in the synovial membrane and cartilage has been suggested as the major cause of osteoarthritis in patients with WD and may contribute to accelerated degenerative changes with deformities, particularly affecting larger joints[99]. In addition, myocardial copper accumulation can cause cardiomyopathy and arrhythmias. Pathological cardiac examination of patients with WD has shown interstitial and replacement fibrosis, intramyocardial small-vessel sclerosis and focal inflammatory cell infiltration[100]. Infrequent presentations include hypoparathyroidism, lunulae, cardiomyopathy, pancreatitis and menstrual irregularities[5,96] (FIG. 3).

Diagnosis, screening and prevention

In WD, copper accumulates pathologically in different organs, causing a wide spectrum of clinical symptoms over the course of WD (FIG. 3), which depend on the organs most affected[1,4,101–103].

Clinical signs and symptoms

Hepatic manifestations. Liver disease is the first clinical manifestation in ~40–60% of patients with WD but may be accompanied by other symptoms[1,104]. In the absence of an established genotype–phenotype correlation[105], symptoms and disease severity may vary among patients and within families. Accordingly, the clinical presentation of WD with liver involvement includes a wide spectrum of signs and symptoms, ranging from asymptomatic subtle morphological changes to the liver and simple acute self-limited hepatitis-like illness to severe hepatitis, recurrent jaundice (in the presence of haemolysis), cirrhosis with or without portal hypertension and even acute liver failure[106]. Age, which affects the duration of untreated copper overload, and sex seem to have a modifying effect on the disease course, as females present more often with acute liver failure than males and adult patients have a higher likelihood of liver cirrhosis than paediatric patients. The exact reason for the varied clinical course of WD is not clear; however, it seems to be multifactorial, and a combination of genetic, epigenetic,

hormonal and environmental factors may have a role. Early diagnosis and treatment are crucial to protect from disease progression and the development of cirrhosis or liver failure[106].

Typically, the first finding in children and young adults with WD is mild to moderate hepatic steatosis, which is evident on liver imaging (by ultrasonography) or by liver biopsy. Hepatic steatosis may be accompanied by abnormal liver function, which can be diagnosed as mildly elevated serum aminotransferases (a marker of liver injury). If patients remain untreated, over time chronic liver disease (fibrosis and cirrhosis) may develop, with complications such as portal hypertension, hepatosplenomegaly, ascites, low serum albumin concentration and coagulopathy. Thus, patients with WD can present both with acute liver failure or chronic liver disease, and WD may be clinically indistinguishable from other hepatic conditions.

The most severe form of hepatic presentation, acute liver failure due to WD (formerly known as 'fulminant WD'), occurs predominantly in young females (the ratio of females to males is 4:1). This state is often associated with Coombs-negative haemolytic anaemia[92], severe coagulopathy, encephalopathy and rapidly progressive renal failure (hepatorenal syndrome). In WD-associated acute liver failure, although serum bilirubin is highly elevated, serum activity of aminotransferases is increased only moderately, and serum concentration of alkaline phosphatase is normal or extremely low; therefore, serum markers of liver injury should be interpreted with caution. The New Wilson Index (modified Nazer score)[107] can offer guidance when evaluating the need for urgent liver transplantation in patients with acute liver failure.

Similar to many other hepatic diseases that may involve cirrhosis, the Model for End-Stage Liver Disease[108] and Child-Pugh scores[109] are commonly used to assess the severity of chronic liver disease. The Model for End-Stage Liver Disease is based on bilirubin, creatinine, the international normalized ratio (INR), sodium and the aetiology of liver disease[108] whereas Child-Pugh scores are based on bilirubin, serum albumin, INR and the degrees of ascites and encephalopathy[109]. Liver imaging by abdominal ultrasonography, CT and MRI are commonly performed at diagnosis. The most frequent findings are fatty infiltration (indicating hepatic steatosis), contour irregularity and right lobe atrophy (both signs of cirrhosis). The role and accuracy of non-invasive measurements of liver stiffness (a marker of chronic liver disease; for example, transient elastography) and biochemical fibrosis scores (based on serum biomarkers of liver injury) in patients with WD remain unclear as only pilot studies in small cohorts are available[110]. For patients with evidence of chronic liver disease, especially in patients with cirrhosis, screening examinations for signs of portal hypertension (oesophagus varices and splenomegaly) and hepatobiliary malignancies[111] should be performed[106] and repeated depending on clinical status (for example, signs of liver failure, ascites, oedema and elevated enzyme levels), although hepatobiliary malignancies are rare.

Patients with WD and decompensated (symptomatic) cirrhosis may present with ascites, jaundice, gastrointestinal bleeding or hepatic encephalopathy. Patients with liver cirrhosis (associated with any aetiology) are susceptible to bacterial infections of any cause, and sepsis is one of the leading causes of death in these patients. Spontaneous bacterial peritonitis is a particular risk and can be diagnosed by paracentesis (testing of ascites fluid). In advanced stages of cirrhosis, there may be complications such as hepatic encephalopathy and hepatorenal syndrome, both of which are associated with high mortality.

Neurological manifestations. After hepatic manifestations, neurological symptoms are the most frequent clinical symptoms of WD. Initial neurological presentation occurs in 18–68% of patients (depending on the referral centre), with mean age at symptom onset of 20–30 years[112]. However, the youngest reported patient with WD with neurological presentation was 6 years old and the oldest was 72 years old[4]. The main clinical spectrum of neurological symptoms includes different movement disorders with a wide variety of involuntary movements, which often overlap[1,4]. Summarizing the most common neurological features of WD, different clinical forms are distinguished where there is a predominance of tremor, dystonia (sustained or repetitive muscle contractions) or parkinsonism (bradykinesia, that is, paucity and slowness of movement, muscle rigidity and resting tremor); all of these movement disorders are often associated with dysarthria (unclear articulation of speech), gait and posture disturbances, drooling and dysphagia (difficulties during any phase of swallowing)[113,114]. These disturbances may severely affect the activities of daily living[86,113,114].

Tremor is a characteristic and frequent neurological symptom in patients with WD, and up to 55% of neurological patients with WD have tremor at diagnosis. Tremor can be resting, postural (often with 'wing-beating' features) or kinetic, may have different frequency and amplitude and may start initially as a unilateral or bilateral tremor, which mainly involves the distal upper extremities. However, as WD progresses, the legs, head or even the whole body may be affected, usually in a bilateral manner. A specific involuntary movement disorder that mimics tremor, called

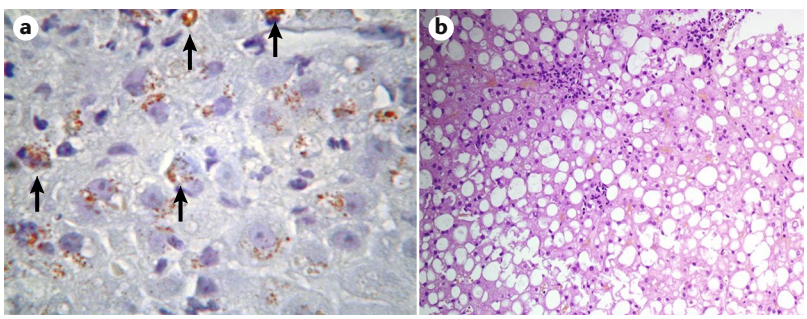


Fig. 4 | **Liver pathology in WD.** Histology performed on liver biopsy samples from a patient with Wilson disease (WD). **a** | Hepatic copper can be observed as rhodanine-positive granules (black arrows). **b** | (Haematoxylin and eosin stain). Early characteristic alterations of the liver pathology in WD include steatosis, which is sometimes indistinguishable from non-alcoholic fatty liver disease. Image **b** adapted with permission from REF.[29], Elsevier.

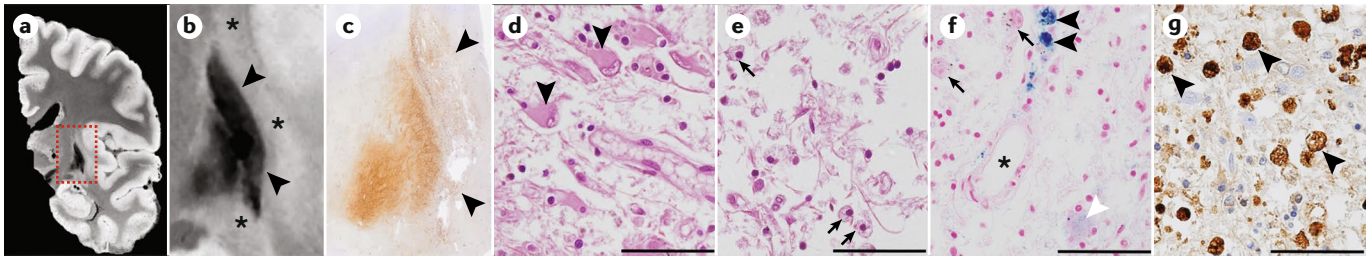


Fig. 5 | **Post-mortem MRI and histopathology in neurological WD.** MRI and histological images taken post-mortem from a patient with neurological Wilson disease (WD). **a** | T_2*-weighted post-mortem MRI acquired with a 7T scanner. **b** | Magnification of the basal ganglia region marked by the dashed red rectangle in part **a**; note the shrunken and markedly hypointense putamen and globus pallidus (marked by black arrowheads) surrounded by a mildly hyperintense area (marked by asterisks). **c** | Low-power magnification of Turnbull iron staining displaying the corresponding area with MRI on image in part **b**; increased iron staining corresponds with abnormally low MRI T_2* signal in parts **a** and **b**. **d**,**e** | Haematoxylin and eosin staining showing reactive astrocytes with large pale nuclei (black arrowheads) and severely damaged neurological tissue with macrophages (black arrows); note that part **d** corresponds to the T_2* hyperintense area directly adjacent to the putamen whereas part **e** corresponds to a rarefied area in the central putamen. **f** | Berlin-blue staining showing iron-negative (black arrows) and iron-positive (black arrowheads) macrophages. Iron is faintly present also in astrocytes (white arrowhead) and as iron dust with dominant perivascular distribution (vessel marked by asterisk); this finding indicates diffusely increased tissue iron deposits. **g** | Ferritin staining shows numerous strongly positive macrophages (black arrowheads) that drive the MRI T_2* signal drop. Of note, the presence of iron-laden macrophages leads to a profoundly decreased T_2* signal even in the putamen, with severe tissue rarefaction. Scale bars in images in parts **d**–**g** represent 50 µm.

asterixis or flapping tremor, is a negative myoclonus affecting the hands that can be observed in patients with WD with liver failure who have developed hepatic encephalopathy[113,114].

Dystonia is reported as the first WD symptom in 11–65% of patients[115,116]. It can be focal (involves one body part, for example, one hand), segmental (involves one body segment, for example, upper extremity), multisegmental (involves multiple segments, for example, face and leg) or may even be generalized (FIG. 6). The most characteristic dystonic presentation of WD is an abnormal facial expression or risus sardonicus, which presents as a fixed smile due to dystonia of the risorius muscle. If WD remains untreated, symptoms usually progress to generalized dystonia, contractures and full immobilization.

Parkinsonism occurs in 19–62% of patients with WD and usually presents as symmetric bradykinesia, rigidity, hypomimia (a reduced degree of facial expression), gait and posture disturbances as well as dysarthria, dysphagia and drooling[86,113,114]. Furthermore, ataxia (impaired coordination of movements) as a symptom of cerebellar dysfunction (ataxic gait and posture, impaired coordination, intentional tremor, dysarthria) occurs in 30% of patients with WD, usually not as a solitary symptom but in combination with other movement disorders[86,114]. Chorea (rapid, irregular involuntary movements of face, head, trunk or extremities) occurs less frequently in patients with WD (6–16%). Gait and posture disturbances are reported in 44–75% of patients with WD with neurological presentation and 57% have impaired hand writing, often an early sign of the disease[116].

Dysarthria appears to be the most frequent neurological symptom and is reported in up to 97% of patients with neurological WD[115,116]. In some cases, dysarthria may be so severe and persistent that verbal communication becomes impossible. Manifestations of dysarthria are not WD-specific but can be divided into cerebellar, extrapyramidal (dystonic and parkinsonian) and mixed (unclassified due to symptoms overlapping)[86,113,114,117].

Dysphagia is reported in up to 50% of neurological patients with WD[86,113,114,118–120]. Dysphagia varies from mild to severe and may lead to severe general health complications, including bronchoaspiration, pneumonia, malnutrition and weight loss. Dysphagia and orofacial dystonia can also lead to drooling, which is observed in almost 68% of neurological patients with WD[114].

In addition to the classical movement disorders of WD, it is notable that other neurological syndromes such as epilepsy may occur[114,121–124]. Current epidemiological studies suggest that epilepsy occurs in 6.2–8.3% of patients with WD, which is a more than tenfold greater frequency than in the general population[89,121]. Seizures are usually generalized across the brain or, less frequently, are focal seizures in a limited area, and seizures can occur during any stage of WD. It has been suggested that additional risk factors for seizures in WD include lesions of white matter tracts in the cortex and may be associated with WD overtreatment and treatment-induced copper deficiency; however, this hypothesis requires further confirmation[115,125].

Other neurological symptoms have been described in patients with WD, including olfactory dysfunction (a suggested marker of neurodegeneration), neuropathy (due to liver failure or treatment-induced copper deficiency), restless leg syndrome, rapid eye movement (REM) sleep behaviour disorder and other sleep abnormalities, tics, myoclonus (involuntary muscle jerks), headache, pyramidal (corticospinal) tract signs, oculomotor impairment and taste dysfunctions; however, there is a lack of studies describing their frequency and relevance[1,4,103,114,115,126–129]. Owing to the wide heterogeneity of symptoms and the combined neurological symptoms occurring in the course of WD, clinical scales were established such as the Unified Wilson's Disease Rating Scale (UWDRS) or the Global Assessment

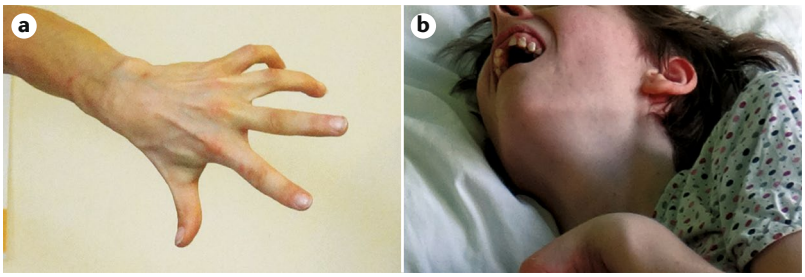


Fig. 6 | **Dystonia, a characteristic symptom in WD.** Dystonia is present in at least one-third of all patients with a neurological presentation of Wilson disease (WD) and can be generalized, segmental, multifocal or focal[1]. Part **a** shows focal hand dystonia. The most characteristic WD dystonic presentation is abnormal facial expression or risus sardonicus, which presents as a fixed smile due to dystonia of the risorius muscle, as shown in part **b**, severe dystonia.

Scale for WD, which assess neurological deficits and functional impairment[4,115,117,130].

Currently, brain MRI is the most important neuroradiological examination for diagnosis of neurological WD and may be helpful for treatment monitoring (discussed below)[4,130]. Usual abnormalities in WD brain MRI include symmetric hyperintense changes visualized in T_2-weighted images located in the basal ganglia (mainly the putamen and caudate nuclei), thalami, midbrain and pons[103,115,131–134] (FIG. 7). In more advanced cases of WD, these abnormalities can be visualized in T_1-weighted images as hypointense changes reflecting severe tissue damage (FIG. 7a,b). The most spectacular brain change associated with WD is described as the 'face of the giant panda' sign in the midbrain (occurring in up to 20% cases of neurological WD) (FIG. 7c), whereas the 'face of the miniature panda' sign occurs less frequently in the pons[115] and injuries of neuronal tracts may also be seen (FIG. 7d). There may be diffuse brain atrophy that is accentuated particularly in the subcortical region and upper brainstem. Tissue atrophy can be present even in patients diagnosed with mild neurological symptoms but is more evident in severe cases (FIG. 7e). In patients with liver failure and/or cirrhosis and portosystemic shunting, an increased signal in T_1-weighted MRI may be observed in the globus pallidus as well as the substantia nigra, which is probably due to manganese accumulation[135] (FIG. 7f,g). Failure of liver detoxification and the presence of portosystemic shunting enables neurotoxic manganese to avoid hepatic metabolism and to enter and accumulate in the central nervous system[136].

Characteristic brain MRI changes are found in almost 100% of treatment-naive patients with neurological WD, 40–75% of patients with hepatic WD and 20–30% of presymptomatic patients. There are no clear correlations between lesion localization and symptoms; however, MRI changes located in the thalamus and pons are suggested to be associated with a poorer prognosis[137]. Very rarely, diffuse white matter changes are observed with preservation of the cortex, probably due to myelin destruction (FIG. 7h). As the presence of MRI brain changes is observed in patients with presymptomatic, neurological or hepatic WD, MRI seems to be justified in all patients before the initiation of therapy. During WD treatment, at least a partial recovery of neuroimaging pathology is usually observed, especially in patients who are diagnosed early in the course of disease[115]; however, not enough data currently exist to justify the use of neuroimaging for routine monitoring. Increased echogenicity may be observed with transcranial ultrasonography in lenticular nuclei and corresponds with changes in MRI, but this method requires specialist skills and is not available at many centres[138].

Ophthalmological manifestations. Pathological copper accumulation in the eyes can cause ophthalmological manifestations of WD, which include the Kayser-Fleischer ring and sunflower cataract[115,139] (FIG. 8). The Kayser-Fleischer ring is caused by copper accumulation in the Descemet membrane (the basement membrane of the corneal epithelium) and appears golden, brown or green in colour at the periphery of the cornea. Kayser-Fleischer rings occur in almost 100% of patients with neurological WD, 40–50% of patients with hepatic WD and 20–30% of presymptomatic patients with WD and as such are an important diagnostic feature[4]; correct identification of Kayser-Fleischer rings requires slit-lamp examination by an experienced observer. Care is needed when using this test as false-positive changes, similar to Kayser-Fleischer rings, are observed in disorders such as primary biliary cirrhosis, cholestasis and neoplastic disorders with high serum copper level (for example, multiple myeloma) and during oestrogen intake (chronic use of oral contraceptives or postmenopausal oestrogen therapy)[103]. Anterior segment optical coherent tomography (OCT), a non-contact ophthalmological procedure that provides images and quantitative analysis of ocular tissue (including cornea), may be an alternative method for detecting copper depositions in the cornea[140] (FIG. 8).

The rarer optical manifestation of WD (occurring in 2–20% of patients), sunflower cataract, appears as a central disk with radiating petal-like fronds located under the lens capsule[90]. The most recent ophthalmologic studies based on OCT and electroretinography suggest that the retina and optic nerve are also affected in WD[90,115].

Psychiatric manifestations. Psychiatric symptoms occur frequently in the clinical presentation of WD[4], mostly secondary to the somatic and brain pathology of the disease. In addition, comorbidity of psychiatric illness (for example, major depressive disorder and bipolar disorder) and WD may also be considered, especially in cases with a strong family history of a particular psychiatric illness.

A 2014 comprehensive review of psychiatric aspects of WD concluded that psychiatric symptoms can occur before, concurrent with or after the diagnosis and treatment of WD[141]. In this review, 20% of patients had seen a psychiatrist before their WD diagnosis, and 30–40% had psychiatric manifestations at the time of diagnosis[141]. Moreover, epidemiological data suggest that up to one-third of patients with WD initially present with psychiatric symptoms[4]. In childhood, a decline in school performance, inappropriate behaviour or impulsiveness may be observed[4]. Some clinical observations also suggest that acute psychiatric symptoms manifest themselves after the initiation of anti-copper agents or in the

first months of treatment, paradoxically even when the neurological status of the patient is improving[142,143].

Mood disturbances are the most common psychiatric manifestation of WD. Between 20% and 60% of patients with WD develop depression, with a high rate of suicidal attempts (4–16% of patients with WD attempt suicide)[14,141,144,145]. The high frequency of depressive syndromes in WD may be associated with a negative patient reaction to diagnosis with a chronic disease as well as the physical incapacity caused by neurological deficits. Around 14–18% of patients with WD have bipolar disorder, which is a higher rate than that seen in the healthy age-matched and sex-matched population[146]. However, studies on the prevalence and incidence of bipolar disorder in patients with WD rarely consider a differentiation between bipolar hypomania or mania and symptoms caused by brain damage. In WD, emotional lability, irritability and aggression, shallow cheerfulness, euphoria, social disinhibition, hypersexuality, lack of criticism and deficits in planning and anticipating social consequences can be associated with lesions of the frontal lobe or its pathways[146].

A few epidemiological studies show that psychosis does not occur more frequently in patients with WD than in the general population; however, psychosis does occur more often in patients with neurological symptoms[141]. Seemingly no specific clinical manifestations of psychosis occur, and patients with WD with psychiatric symptoms usually have an initial diagnosis of schizophrenia, schizoaffective and delusional disorder. Psychotic symptoms that occur as the first manifestation of WD may present both diagnostic and therapeutic challenges[147]. As ~3% of first-episode psychosis may be caused by 'organic' causes (for example, copper toxicity in WD), some diagnostic guidelines suggest screening for WD in first-episode psychotic patients[148,149].

Behavioural and personality disorders are also frequent psychiatric disturbances seen in patients with WD, and the most common manifestations are irritability, aggression and antisocial behaviour[150]. Other psychiatric conditions such as catatonia, anorexia nervosa, bulimia, obsessive-compulsive disorder and attention-deficit/hyperactivity disorder have also been reported in WD[151,152]. Unfortunately, in most described cases, psychiatric manifestations lead to a delay in the diagnosis of WD.

Establishing the diagnosis

As described above, clinical presentation varies widely in patients with WD and, in most cases, a combination of clinical features and laboratory parameters (TABLE 1) are required to establish the diagnosis.

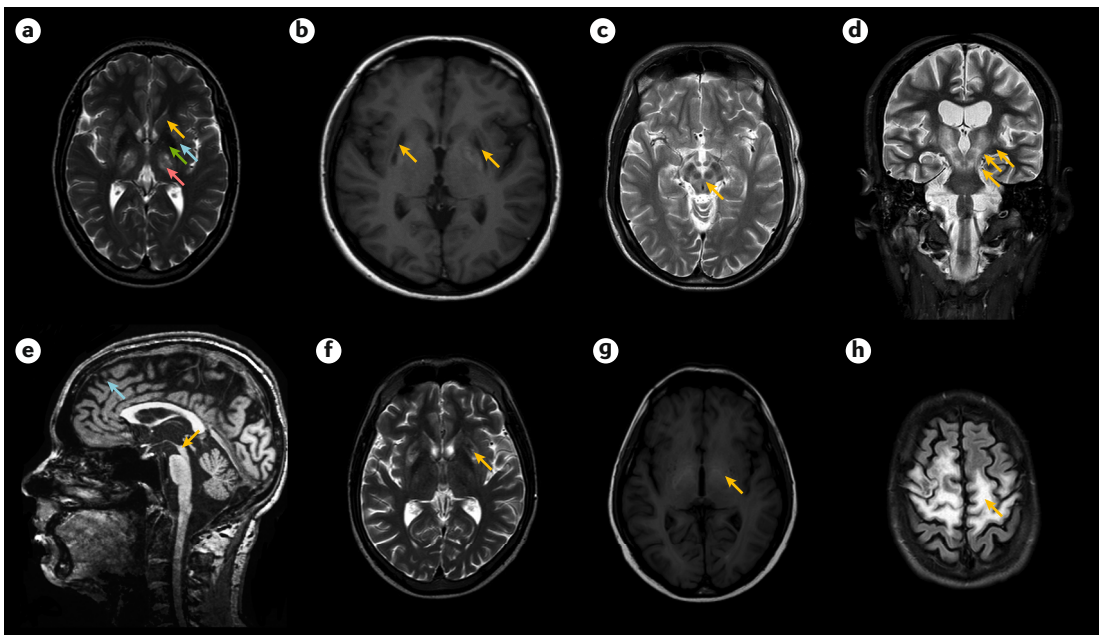


Fig. 7 | **Brain MRI changes in WD.** Usual abnormalities in brain MRI in patients with Wilson disease (WD) include symmetric hyperintense changes visualized in T_2-weighted images of the basal ganglia, particularly the putamen (blue arrow), caudate nuclei (orange arrow), thalami (pink arrow) and globi pallidi (green arrow) (part **a**). In more advanced cases, severe tissue damage can be visualized in T_1-weighted images as hypointensity in the putamen (part **a**, blue arrow; part **b**, orange arrows). The most spectacular changes observed in WD are described as the 'face of the giant panda' in the midbrain (part **c**, orange arrow). Another common MRI abnormality is increased T_2 signal along the dentato-rubro-thalamic pathway (part **d**, orange arrow), which is a major efferent pathway from the cerebellum involved in movement disorder symptoms, including ataxia, tremor and dystonia. In severe neurological WD, diffuse brain atrophy (part **e**) may be seen in the midbrain (orange arrow) and cortex (blue arrow). In brain MRI scans from a 21-year-old male with severe liver failure and discrete neurological signs, hyperintense changes in putamina can be visualized in T_2-weighted images, which is characteristic of early stages of brain involvement (part **f**, orange arrow). Hyperintense changes in the globi palidi in T_1-weighted images in the same patient are presumably due to manganese accumulation, which is characteristic of hepatic encephalopathy (part **g**, orange arrow). Very rarely, diffuse white matter changes in both brain hemispheres are observed with preservation of the cortex, probably due to myelin destruction (part **h**, orange arrow).

Serum levels of ceruloplasmin can be tested if WD is suspected. Serum ceruloplasmin is typically decreased in patients with neurological WD compared with healthy individuals but may be in the low to normal range in about half of patients with active liver disease[153]. As such, the predictive value of ceruloplasmin for diagnosis of WD in patients with liver disease is poor[154,155]. Serum ceruloplasmin levels can be measured enzymatically or by nephelometric immunoassays. However, immunological assays may overestimate ceruloplasmin concentrations because they do not discriminate between apoceruloplasmin and holoceruloplasmin[156].

Total serum copper is usually decreased in proportion to reduced levels of ceruloplasmin (holoceruloplasmin and apoceruloplasmin) in the circulation. Normal or elevated serum copper levels with decreased levels of ceruloplasmin indicate an increase in the concentration of non-ceruloplasmin-bound copper, which is suggestive of WD. The concentration of non-ceruloplasmin-bound copper can be estimated by subtracting ceruloplasmin-bound copper from the total serum copper concentration[157]. However, the calculation of non-ceruloplasmin-bound copper concentration depends on the correct determination of serum levels of functional ceruloplasmin, which is more precisely addressed when using enzymatic assays. Exchangeable copper is an experimental technique that aims to determine bioavailable copper in the plasma compartment[158]; however, this technique does not reliably measure non-ceruloplasmin-bound copper concentration.

A useful measure is 24-hour urinary copper excretion, which is helpful to diagnose WD and to monitor treatment. The conventional level taken as diagnostic of WD is >100 µg excreted over 24 h (>1.6 µmol per 24 h) in patients with signs and symptoms of WD[4,5]. However, basal 24 h urinary copper excretion may be <100 µg at presentation in 16–23% of patients diagnosed with WD, especially in children and asymptomatic siblings[4,5]. Moreover, the interpretation of 24 h urinary copper excretion values can be difficult owing to overlap with findings from other types of liver disease, in particular acute liver injury or liver failure of any aetiology. Furthermore, the reference limits for normal 24 h excretion of copper vary among clinical laboratories. Many laboratories use 40 µg per 24 h (0.6 µmol per 24 h) as the upper limit of normal, which appears to be a better threshold for diagnosis[5].

A greater than fivefold increase in urinary copper excretion after challenge with oral D-penicillamine (a chelator) has been used for diagnostic purposes in children if the measurement of 24 h urinary copper excretion alone is inconclusive[159]. Although this test is used in some centres, its diagnostic value is not confirmed[6,160]. Moreover, this test may not be required if the alternative lower threshold for urinary copper excretion of 40 µg per 24 h is applied. A radioactive copper test, which shows incorporation of copper into ceruloplasmin, may be used when standard copper metabolism tests and genetic tests are inconclusive (for example, to distinguish heterozygotes from homozygotes)[161]; however, this test is available only in highly specialized centres.

If WD is suspected, a liver biopsy with measurement of hepatic parenchymal copper concentration is required if the clinical signs and non-invasive tests do not allow a final diagnosis or if there is suspicion of additional liver pathology from other causes[4]. A hepatic copper content of >250 µg (4 µmol) per gram of dry liver weight is considered robust biochemical evidence for WD. In a large prospective study, 209 µg (3.3 µmol) per gram dry weight provided the highest diagnostic accuracy for WD[162] (sensitivity of 99.4% and specificity of 96.1%). However, hepatic copper content can be underestimated owing to sampling errors[163,164]. Of note, hepatic copper content may also be increased in cholestatic disorders[163].

Mutational analysis is a further important diagnostic tool. However, the results of direct molecular-genetic diagnosis may take time to obtain as analysis is difficult owing to the number (>700) of possible mutations[16,165,166] and as many patients are compound heterozygotes. However, sequencing is becoming continually faster and less expensive; therefore, it is likely to become a commonly used diagnostic test in the future.

None of the available laboratory tests are completely accurate and specific for WD, and typical clinical

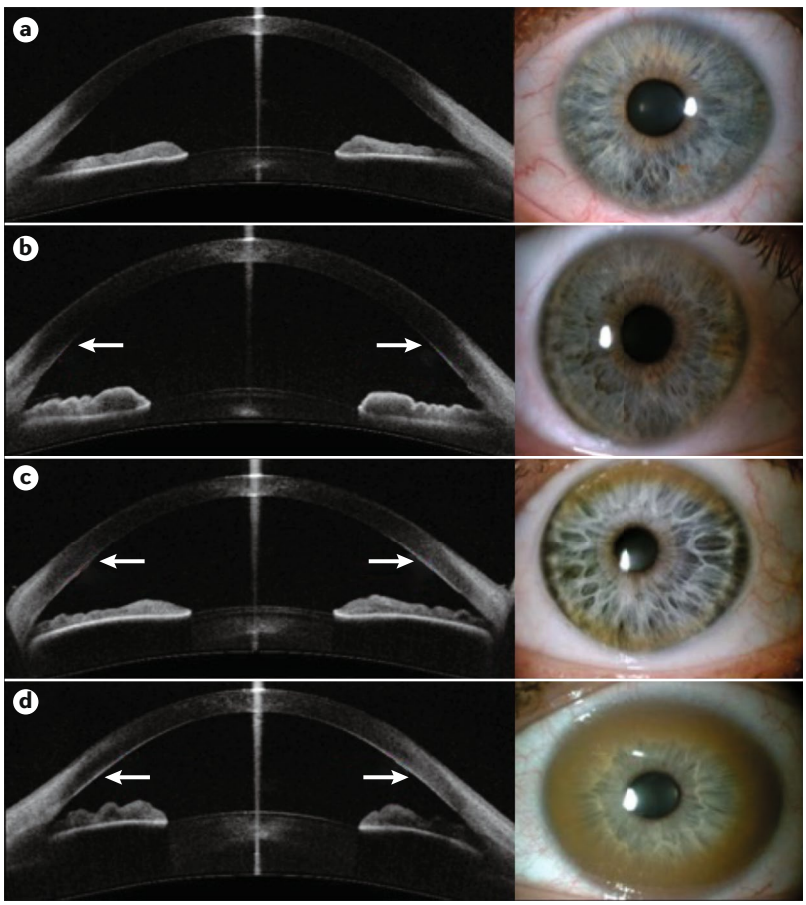


Fig. 8 | **Kayser-Fleischer rings in WD.** Anterior segment optical coherent tomography (OCT) of the eye in healthy individuals and patients with Wilson disease (WD). In some patients with WD and in healthy individuals, corneal copper deposits are not seen (part **a**). However, in a substantial proportion of patients with WD, copper deposits can be visualized by anterior segment OCT as hyper-reflective points that are discrete (part **b**), on the superior and inferior part of the cornea (part **c**) or in the formation of a complete Kayser-Fleischer ring (part **d**). Images courtesy of K. Broniek and J. Szaflik, the Department of Ophthalmology, Medical University of Warsaw, Warsaw, Poland.

Table 1 | **Routine tests for the diagnosis of Wilson disease**

Test	Typical finding	False 'negative'	False 'positive'
Serum ceruloplasmin	Decreased in patients with WD compared with healthy controls	• Normal levels in patients with marked hepatic inflammation • Overestimation by immunological assay • Pregnancy • Oestrogen therapy	Low levels in patients with malabsorption, malnutrition and/or aceruloplasminemia and in heterozygotes
24 h urinary copper	• Adults: >100 µg (1.6 µmol) per 24 h • Child: >40 µg (0.64 µmol) per 24 h	• Normal levels caused by incorrect collection • Normal levels in children without liver disease	• Increased in hepatocellular necrosis • Increased in cholestasis • May appear increased owing to sample contamination
Non-ceruloplasmin-bound copper	>10 µg dl^{-1} (1.6 µmol per litre)	May appear normal or negative if ceruloplasmin is measured by immunological assay	NA
Hepatic copper	250 µg (4 µmol) g^{-1} dry weight	• Regional variation in patients with active liver disease • Regional variation in patients with regenerative nodules	• Increased in cholestatic syndromes • Increased in idiopathic copper toxicosis disorders
Kayser-Fleischer rings by slit-lamp examination	Present	• Absent in up to 50% of patients with hepatic WD • Absent in most asymptomatic siblings	May be present in primary biliary cholangitis (primary biliary cirrhosis)

NA, not applicable; WD, Wilson disease. Adapted with permission from REF.[4], Elsevier.

symptoms may often be absent; therefore, a diagnostic score based on all available tests was proposed by the Working Party at the Eighth International Meeting on Wilson Disease, Leipzig 2001[167] (TABLE 2). Importantly, the score was found to have good diagnostic accuracy and was adopted in the European Association for the Study of the Liver (EASL) Clinical Practice Guidelines for WD[4].

Prevention

WD is one of the few metabolic disorders that can be successfully pharmacologically treated if diagnosis is established early. When patients with WD who are presymptomatic are treated, they have similar survival to the general population, providing they have good adherence to treatment[3]. Earlier diagnosis can be achieved by the family screening of first-degree relatives of patients with WD and the differential diagnosis of other liver and movement disorders, thereby allowing the disease to be correctly treated. It should be emphasized that one of the most important factors that determines poor treatment outcome is delayed diagnosis[4,5,167,168]. Individuals who are at the greatest risk of WD are siblings of an index case; in this case, to prevent further symptomatic disease, obligatory fast diagnosis is needed regardless of age, even if there is a lack of symptoms[169]. The long-term follow up of a large cohort of 760 Polish patients suggests that the children of patients with WD have a ~4% risk of developing WD. There is also an increased risk of the disease among cousins and parents; WD has been diagnosed in asymptomatic parents of patients with WD, and screening of the previous generation is useful[169,170]. Patients must be informed about the higher risk of WD in family members, especially when liver, neurological or psychiatric symptoms occur. Genetic screening is not always available or conclusive, and the normal ranges of copper metabolism parameters are not well established in small children; therefore, diagnosis of WD must be supported by careful clinical observation. In small infants, it is not clear when anti-copper therapy should be started to avoid the consequences of copper deficiency. Knowledge about WD at all stages among patients and their families is very important, and there is a valuable place for patient support organizations[171].

Management

If a diagnosis of WD is established, treatment should always be lifelong[4,5,167]. Current management options include pharmacological therapies and liver transplantation[115,172]. To summarize management, first, WD may be successfully treated with pharmacological agents if diagnosed early (before advanced liver or neurological damage) and lifelong adherence to treatment is maintained; second, the drug choice should be discussed with patients on the basis of possible adverse events, availability and costs; third, treated patients with WD should undergo continued monitoring to assess adherence to treatment, including copper status, liver function, neurological and psychiatric status and any effects of the disease on other organs; and fourth, symptomatic treatment of liver, neurological or psychiatric manifestations should be applied if needed and reassessed over time.

Pharmacological treatment

The pharmacological treatment of WD is based on drugs that induce a negative copper body balance and includes chelators (D-penicillamine and trientine) that increase urinary copper excretion and zinc salts that decrease copper absorption from the digestive tract[13–15,173,174]. With correct and early pharmacological treatment (before the development of advanced liver disease (such as cirrhosis) or brain injury), markers of liver function and injury improve in >90% of patients, usually in 2–6 months, whereas clinical neurological improvement is observed in ~50–60% of patients over a longer time course of 1–3 years[4]. Adherence has an extremely important role for the long-term success of treatment, both in symptomatic and presymptomatic patients with WD[3,175].

Table 2 | **Diagnostic scoring system for Wilson disease**[167]

Test	Parameter	Score
Typical clinical signs and symptoms		
Kayser-Fleischer rings	Present	2
	Absent	0
Neurological symptoms[a]	Severe	2
	Mild	1
	Absent	0
Serum ceruloplasmin	Normal (>0.2 g per litre)	0
	0.1–0.2 g per litre	1
	<0.1 g per litre	2
Coombs-negative haemolytic anaemia	Present	1
	Absent	0
Other tests		
Liver copper (in the absence of cholestasis)	>250 µg (>4 µmol) g^{-1} dry weight	2
	50–249 µg (0.8–4 µmol) g^{-1}	1
	Normal: <50 µg (<0.8 µmol)	−1
	Rhodanine-positive granules[b]	1
Urinary copper (in the absence of acute hepatitis)	Normal	0
	1–2 × ULN	1
	>2 × ULN	2
	Normal but >5 × ULN after D-penicillamine	2
Mutation analysis	On both chromosomes detected	4
	On one chromosome detected	1
	No mutations detected	0
Total score	***Evaluation***	
≥4	Diagnosis established	
3	Diagnosis possible; more tests needed	
≤2	Diagnosis very unlikely	

ULN, upper limit of normal. [a]Or typical abnormalities at brain MRI. [b]If no quantitative liver copper available. Adapted with permission from REF.[167], John Wiley & Sons.

In patients with WD undergoing pharmacological therapy, monitoring should be performed at least twice a year to determine the effectiveness of treatment with regards to clinical improvement and biochemical changes (for example, urinary copper excretion and non-ceruloplasmin-bound copper levels), to assess treatment adherence and to detect any treatment-induced adverse events[5]. An acute clinical presentation with rapid deterioration may occur in patients with WD who stop their medication. Failure to adhere to lifelong therapy can lead to substantial progression of WD-associated liver disease and/or liver failure, the latter requiring liver transplantation for survival[4,5]. Studies suggest that up to 45% of patients who are treated with current pharmacological therapies have poor or problematic long-term adherence[169,175]; therefore, adherence should be carefully monitored in patients with WD with all forms of disease presentation[4,5].

For patients on copper chelation therapy, elevated values in the urinary copper test suggest nonadherence to treatment and hepatic deterioration may follow. Low values for urine copper excretion for patients who are on chelation treatment can also indicate overtreatment, and this finding may be accompanied by very low values for estimates of non-ceruloplasmin-bound copper. Moreover, neutropenia, anaemia and hyperferritinaemia (high blood levels of the iron storage protein ferritin) may also indicate copper deficiency. Data regarding WD drug interactions, treatment monitoring, effectiveness and adherence assessment as well as adverse events are presented in TABLE 3.

An important adverse event that can occur with each type of treatment (including liver transplantation) is paradoxical neurological deterioration, with rapid appearance of new neurological signs and symptoms or worsening of existing neurological signs[137]. Paradoxical neurological deterioration with the first available chelator, D-penicillamine, prompted the discussion whether it should be used in patients with neurological WD and led to the search for safer treatments[101,176]. The mechanism of paradoxical neurological deterioration is still not proven but may be caused by the rapid mobilization of copper from different tissues, upon chelator initiation, with a subsequent secondary increase of non-ceruloplasmin-bound copper into the blood, which then causes increased oxidative stress and cell damage in the brain[4,115,177]. On the basis of this model, chelators are now introduced slowly with dose titration, and the occurrence of paradoxical neurological deterioration with different treatments seems similar (~10%) and lower than observed initially with D-penicillamine[124,160,162]. Other suggested risk factors for paradoxical neurological deterioration include additional drugs (for example, neuroleptics and antidepressants) and advanced neurological and brain MRI changes[13,137]. Additionally, it is sometimes impossible to distinguish paradoxical neurological deterioration from the natural course of the disease, especially in late-diagnosed cases[13,137].

The current guidelines from EASL[4] and the American Association for the Study of Liver Diseases (AASLD)[5] recommend the use of chelators as the initial treatment for all patients with symptomatic WD, with the suggestion that trientine is better tolerated. Owing to relatively slow effects, zinc salts are generally reserved for maintenance treatment (after biochemical and clinical improvement), although they are also used as first-line therapy, most commonly for asymptomatic or presymptomatic patients[4,5]. First-line zinc monotherapy appears to be effective and well tolerated in patients with neurological symptoms, and paradoxical neurological deterioration is uncommon[4]; however, caution is needed in patients with hepatic WD owing to the risk of hepatic deterioration that has been occasionally reported[4]. The use of zinc monotherapy in patients with mild liver disease (based on liver aminotransferases) that is diagnosed in childhood is supported by retrospective data[178].

It should be emphasized that there are very limited data from prospective head-to-head clinical trials that compare the safety and efficacy of different pharmacological treatment options in WD[4,5,115]. There are conflicting data from retrospective studies according to the superiority or equality of different drugs, which is mainly based on centre-specific or country-specific experience with different drugs[4,5,13–15,115,145,173,174,178,179].

Liver transplantation

A surgical and more complex option for the treatment of WD is liver transplantation[107,172,180]. As WD is primarily a disease of the liver characterized by defects in copper metabolism in hepatocytes, liver transplantation can be considered a phenotypical correction of the gene defect and can restore copper homeostasis. According to EASL and AASLD recommendations[4,5], liver transplantation in patients with WD is indicated in acute liver failure or decompensated liver cirrhosis, which does not respond to pharmacological treatments of WD. A transplant indication in the setting of acute liver failure is usually based on a specific WD clinical scale, commonly the revised King's prognostic Wilson Index[107] (TABLE 4). Whether uncontrolled neurological disease constitutes an indication for liver transplantation, as a 'last-chance' treatment option, is highly controversial[107,172,180]. Notably, paradoxical neurological deterioration may be observed in liver transplantation patients; however, the mechanism is unclear. Acute liver failure is often associated with a rapid increase of copper in blood, causing acute haemolysis and allowing copper penetration to the brain. Anaesthetics used during liver transplantation surgery may also induce neurological deterioration.

Symptomatic therapies

Dietary copper restriction may be helpful in controlling copper excess, but dietary management is not recommended as sole therapy. Patients with WD should generally avoid foods with very high concentrations of copper (for example, shellfish, nuts, chocolate, mushrooms and organ meats) at least in the first year of pharmacological treatment[5,50].

In addition to specific anti-copper treatments, symptomatic therapies are important in the management of WD, particularly to address complications of liver injury or failure[181] as well as neurological symptoms[182]. In patients with documented liver disease (hepatic steatosis, liver fibrosis or cirrhosis), symptomatic management may include the avoidance of further liver injury caused by the intake of potentially hepatotoxic substances (for example, alcohol, herbal supplements and drugs) and screening and/or re-evaluations for oesophageal or gastric varices[181] and hepatocellular carcinoma. In the case of decompensated liver cirrhosis, symptomatic management also includes treating the complications of portal hypertension, which are gastroesophageal varices, ascites, hepatic encephalopathy, spontaneous bacterial peritonitis and hepatorenal syndrome. Symptomatic liver therapy should always be introduced in accordance with good hepatology practice without any delay while waiting for a response to anti-copper treatment effects[181].

Symptomatic treatment should also be considered in patients with WD with persistent or deteriorating neurological symptoms during treatment[182]. The symptomatic treatment of neurological impairments depends mainly on the predominant symptoms, such as dystonia, parkinsonism or tremor, and is based on experiences from treatment in other neurodegenerative diseases. No symptomatic treatments for neurological WD have been tested in clinical studies. Moreover, it is difficult to distinguish a positive effect of symptomatic treatments from an improvement in symptoms due to anti-copper

Table 3 | **Drugs used in the treatment of Wilson disease**

Drug	Mode of action	Interactions	Frequency of AEs leading to treatment discontinuation	Assessment of treatment effectiveness and adherence
DPA	Promotes urinary excretion of copper	• Do not combine with myelosuppressive agents, cytostatic agents, antimalarials, gold therapy, oxyphenbutazone or phenylbutazone • DPA interacts with heavy metals	• 20–30% during treatment[174,179] • 'Early' AEs (first 3 weeks) include fever, cutaneous manifestations, lymphadenopathy, arthralgia, leukopenia, thrombocytopenia and proteinuria • 'Late' AEs (3 weeks to years) include paradoxical neurological worsening, renal insufficiency, fatal glomerulonephritis, intra-alveolar haemorrhage, myasthenia-like syndrome, lupus-like syndrome, fatal bone marrow aplasia, gastric symptoms, hair loss or loss of taste	• Copper urinary excretion 200–500 µg per 24 h (at the beginning of the treatment >1,000 µg per 24 h) • Serum NCC[a] 5–15 µg dl^{-1} • Normalization of copper urinary excretion 2 days after stopping the treatment with DPA
Trientine	Promotes urinary excretion of copper	• Mineral supplements should be avoided	• 7.1%[179] • Gastritis • Sideroblastic anaemia • Lupus-like reactions • Loss of taste	• Copper urinary excretion 200–500 µg per 24 h (at the beginning of the treatment >1,000 µg per 24 h) • Serum NCC 5–15 µg dl^{-1} • Normalization of copper urinary excretion 2 days after stopping the treatment with trientine
ZSs	Blocks intestinal absorption of copper	• ZSs diminish absorption of tetracyclines and quinolones • Diuretics increase urinary zinc excretion • Iron salts, milk, milk products, whole-grain bread, products containing phytates, high-fibre foods, and chelating agents diminish absorption of ZSs	• 3–7%[174] • Gastritis • Biochemical pancreatitis • Immunosuppression • Bone marrow depression	• Copper urinary excretion <75 µg per 24 h • Serum NCC 5–15 µg dl^{-1} (>12 months of treatment)

AE, adverse effect; DPA, D-penicillamine; NCC, non-ceruloplasmin-bound copper; ZSs, zinc salts. [a]NCC is not routinely used for monitoring treatment efficacy but has been used as an efficacy measure in a clinical trial of tetrathiomolybdate[199].

Table 4 | **Wilson disease prognostic index**

Parameter	0 points	1 point	2 points	3 points	4 points
Serum bilirubin (micromoles per litre)	0–100	101–150	151–200	201–300	>301
Aspartate aminotransferase (units per litre)	0–100	101–150	151–300	301–400	>401
International normalized ratio	0–1.29	1.3–1.6	1.7–1.9	2.0–2.4	>2.5
White blood cell count (10^9 per litre)	0–6.7	6.8–8.3	8.4–10.3	10.4–15.3	>15.4
Albumin (grams per litre)	>45	34–44	25–33	21–24	<20

A score of ≥ 11 points is associated with high probability of death without liver transplantation and is an indication for liver transplantation. Adapted with permission from REF.[107], John Wiley & Sons. New Wilson Index modified from the Nazer scoring system[210] by Dhawan et al.[107].

therapy[13]. In patients with severe neurological WD, there is often either a weak or no effect with symptomatic treatments, such as dopaminergic or anticholinergic medication. Tube feeding may be important in cases of severe dysphagia, and percutaneous gastrostomy should be discussed to avoid bronchopneumonia and malnutrition. Drooling may be successfully treated with botulinum toxin injections in salivary glands[182].

Management during pregnancy

In women, amenorrhoea and frequent miscarriage may precede the onset of symptomatic WD and are more frequent in patients with symptomatic WD than in the general population[183–185]. Aside from the increased risk of miscarriage, pregnancy in patients with WD with compensated liver disease is safe, and most patients will have successful pregnancies. The management of anti-copper therapy during pregnancies should focus on the prevention of spontaneous miscarriage, the control of maternal WD and minimization of putative drug-induced teratogenicity, with dose reductions of chelators advisable as a cautionary measure owing to a lack of data. Pre-conception counselling might address the risks of anti-copper medication for the pregnancy, but it may also increase the risk of uncontrolled disease as women may choose to not take anti-copper medication during the pregnancy[183,185,186].

In the case of successful conception, a biochemical and clinical baseline assessment of the mother is mandatory, including the appraisal of portal hypertension in patients with cirrhosis (who have an increased risk of peripartum variceal haemorrhage), which will enable a delivery plan to be made[187]. As prevention of symptomatic deterioration of the mother is the primary concern, there is no rationale for discontinuation of any anti-copper therapy during pregnancy, which may lead to severe liver or neurological exacerbation[183]; however, dose reduction may prevent copper deficiency, with an adjustment to account for increased fetal copper demand[188].

For patients on a chelator (D-penicillamine or trientine), a reduced daily dose might be appropriate during the first and second trimester. At the beginning of the third trimester, on an individual basis (taking into account the outcome of biochemical liver function tests during pregnancy), a further daily dose reduction can be considered[186]. After delivery, up-dosing of the chelating agents to the pre-pregnancy level should be considered. There is no evidence that switching the medical therapy to zinc before conception decreases the risk of miscarriage or occurrence of birth defects[186]. From clinical experience, in patients treated with zinc, the daily dose of elementary zinc can be maintained during pregnancy in almost all cases[186]. As all available anti-copper drugs pass into breast milk and may cause infantile copper deficiency, breastfeeding is not generally recommended[5,188].

Management of psychiatric symptoms

In the management of psychiatric disturbances in WD, there should be a consideration of the limitations of psychoactive drugs owing to WD-associated liver disease and the potential effect of drugs on worsening neurological signs[149]. Selective serotonin reuptake inhibitors (SSRIs) may be used as a first-line treatment for depression. In addition, serotonin-noradrenaline reuptake inhibitors, serotonin antagonist and reuptake inhibitors and electroconvulsive therapy (ECT) have been used in patients with WD with good effect[189,190]. Antidepressants with high risk of liver injury, such as iproniazid, phenelzine, imipramine, amitriptyline, duloxetine, bupropion or agomelatine, should be avoided[4,191]. For treatment of hypomania or mania, mood stabilizers may be used, such as lithium and antiepileptic drugs (for example, carbamazepine and lamotrigine, but valproate is avoided owing to hepatotoxicity)[146,192].

Antipsychotic drugs are used in severe mania (in patients with bipolar disorder) and for the treatment of psychotic symptoms. However, in patients with WD, they pose a risk of neurological deterioration and hepatic injury. Therefore, antipsychotics with low risk of extrapyramidal symptoms, such as clozapine or quetiapine[193], should be used. Clozapine should be reserved for the most severe and treatment-resistant cases owing to the increased risk of leukopenia and dose-related seizures, with regular blood analysis[4]. Olanzapine is an effective anti-manic drug with low extrapyramidal risk[146], whereas olanzapine and quetiapine have a moderate risk in patients with liver injury[191]. In addition, aripiprazole has a good safety profile and sulpiride and amisulpride are interesting options because they are not metabolized in the liver and have a low risk of extrapyramidal symptoms, especially at lower doses. Antipsychotic drugs, even those considered as having low effect on the extrapyramidal system, should be used only in severe cases, at the lowest effective dose and for the shortest possible duration[114,130,149].

Finally, psychiatric treatment of obsessive-compulsive disorder should include SSRIs with cognitive behavioural therapy, and lorazepam followed by ECT should be considered for catatonia[189]. For behavioural symptoms and personality disorders, SSRIs, antiepileptics and benzodiazepines may be used[149].

Quality of life

Quality of life (QOL) is one of the most important objective patient-reported outcomes of the treatment of chronic disorders; however, QOL has not been clearly investigated in WD. Only four studies (only one with liver-transplanted patients with WD) have been

performed, which aimed to verify the 36-item Short Form Health Survey (SF-36) and the WHO QOL Brief Questionnaire (WHOQOL-BREF) in patients with WD[194–197]. The main conclusion from these studies is that patients who experience a long delay before receiving WD treatment have poor QOL[194], which highlights the importance of early WD diagnosis. Owing to the wide spectrum of symptoms, the non-homogeneous clinical presentation and the limited numbers of studies (none with drug-naive patients; most without healthy control groups), it is difficult to recommend any QOL scale (or their subscales) to use routinely in WD. Additionally, it should be noted that large cohorts of patients with WD that have associated psychiatric disturbances, including cognitive, behavioural or criticism disturbances, may decrease the reliability of assessment (authors' unpublished data). Further studies are needed, based on larger groups of patients, as well as including the objective correction of self-reported QOL scales, before QOL scales may be used in the routine assessment of patients with WD.

Outlook

Improved diagnostic techniques and more systematic approaches to the diagnosis of WD have created an easier path to confirmation or exclusion of a diagnosis of WD. Available therapies of chelating agents and zinc are mostly effective for the management of WD but have some limitations[19,198]. A need exists for improved information about dosing, patient monitoring, studies of the comparative effectiveness of the available medical therapies and standardized definitions for treatment success and failure. A recent phase II trial in 28 patients with WD with tetrathiomolybdate, which is an agent that leads to the formation of an inert complex of tetrathiomolybdate with copper and albumin in the circulation and increases biliary copper excretion, seems very promising, especially with respect to the low potential for neurological worsening with the initiation of therapy[199]. Of note, owing to the lifelong nature of WD treatment, new therapies must be accessible and affordable if they are to be clinically useful, particularly when health-care resources are stretched and as regulatory authorities consider WD a rare disease.

New advances in molecular genetics may soon enable the clinical translation of successful studies of gene therapy using viral vectors with *ATP7B* constructs in animal models of WD to patients with WD, which may potentially provide a 'cure' for this disorder[200]. Future applications of somatic gene modification (such as CRISPR–Cas9 genome editing) may permit the correction of some *ATP7B* mutations by restoring functional copper transport in liver cells. Hepatocyte cell transplantation may offer another modality for treatment to correct copper metabolism in WD, but it requires immunosuppression to prevent rejection of thetransplanted cells and may not necessarily correct the complications of portal hypertension as accomplished by liver transplantation. Future manipulation of autologous stem cells may allow their transformation into functional liver cells, thereby eliminating the need for subsequent immunosuppression to maintain transplanted cells. Other treatments may focus on the ability to restore some function to certain mutant ATP7B proteins by changing their conformation or intracellular localization[201]. These novel therapies may offer the future opportunity to lower or even discontinue the current standard-of-care treatments for WD.

Despite current advances, there are still unmet needs for patients with WD and other unexplained issues regarding the wide phenotypic expression of disease. The development of neonatal screening techniques that can detect most patients with WD before the development of symptoms is an important priority[202]. To increase the understanding of phenotypic variation, the complex interrelationships between environmental and host factors need to be further explored[203]. Some studies have shown that genetic and epigenetic factors influence whether neurological disease may progress differently in certain individuals or whether these individuals develop hepatic steatosis or metabolize copper differently, all independent of mutations in *ATP7B*. Other studies in animal models have shown that nutritional changes can induce changes in gene expression by alterations in DNA modification[204]. A deeper understanding of these factors along with more careful phenotypic characterization of patients will help us to explore whether we can individually predict the natural history of the disease and response to therapy. In addition, new neuroimaging techniques such as magnetic resonance quantitative susceptibility mapping, diffusion tractography and PET may, in the future, broaden our knowledge regarding in vivo brain pathology in WD and help with prognostication after treatment[83,131,205,206].

Overall, the outlook for patients with WD is bright, for never in the history of WD have there been as many opportunities for the early diagnosis of disease and implementation of effective medical therapies or the availability of rescue therapies, such as liver transplantation. New opportunities for improving diagnosis and treatment of WD will augment those in use now, and even better outcomes may be achievable.

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Competing interests
A.C. has served on advisory boards for Wilson Therapeutics, Vivet Therapeutics and GMP-Orphan SAS and has received speaker fees from EVER Pharma, Boehringer Ingelheim and Nutricia. P.F. has served on advisory boards for Wilson Therapeutics, Vivet Therapeutics and Univar and has received speaker fees from Univar. V.M. has served as a consultant for Kadmon Holdings. K.H.W. is on the speakers bureaus of AbbVie, Alexion Pharmaceuticals, Bayer, Bristol-Myers Squibb, Chiesi Farmaceutici SpA, GMP-Orphan SAS, Norgine, Novartis, Univar, Wilson Therapeutics and Vivet Therapeutics and has received grants (to the institution) from Alexion Pharmaceuticals, Bayer, Bristol-Myers Squibb, Eisai, GMP-Orphan SAS, Novartis, Univar and Wilson Therapeutics. M.L.S. has served on advisory boards for Wilson Therapeutics, Vivet Therapeutics, GMP-Orphan SAS and Kadmon Holdings, is a speaker for Gilead Sciences and is on the Medical Advisory Committee of the Wilson Disease Association. All other authors declare no competing interests.

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