Trichothiodystrophy: Update on the sulfur-deficient brittle hair syndromes

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Trichothiodystrophy (TTD) refers to a heterogeneous group of autosomal recessive disorders that share the distinctive features of short, brittle hair and an abnormally low sulfur content. Within the spectrum of the TTD syndromes are numerous interrelated neuroectodermal disorders. The TTD syndromes show defective synthesis of high-sulfur matrix proteins. Abnormalities in excision repair of ultraviolet (UV)-damaged DNA are recognized in about half of the patients. Three distinct autosomal recessive syndromes are associated with nucleotide excision repair (NER) defects: the photosensitive form of TTD, xeroderma pigmentosum, and Cockayne syndrome. The unifying feature of these conditions is exaggerated sensitivity to sunlight and UV radiation. In contrast to patients with xeroderma pigmentosum, no increase of skin cancers in patients with TTD has been observed. Genetically, 3 complementation groups have been characterized among photosensitive patients with TTD. Most patients exhibit mutations on the two alleles of the XPD gene. Rarely, mutated XPB gene or an unidentified TTD-A gene may result in TTD. In UV-sensitive TTD, the TFIIH transcription factor containing XPB and XPD helicase activities necessary for both transcription initiation and DNA repair is damaged. Beyond deficiency in the NER pathway, it is hypothesized that basal transcription may be altered leading to decreased transcription of specific genes. Depressed RNA synthesis may account for some clinical features, such as growth retardation, neurologic abnormalities, and brittle hair and nails. Therefore the attenuated expression of some proteins in differentiated cells is most likely explained by a mechanism distinct from DNA repair deficiency. The first transgenic mouse models for NER deficiencies have been generated. The TTD mouse as well as related cell models will provide important tools to understand the complex relationships between defects in DNA repair, low-sulfur hair shaft disorders, and the genotype-phenotype correlates for this constellation of inherited disorders, including the lack of predisposition to cancer in patients with TTD. (J Am Acad Dermatol 2001;44:891-920.)

Learning objective: At the completion of this learning activity, participants will have a current understanding of the expanded and further defined clinical spectrum of the TTD syndromes. Participants will have gained new insight into the genetic and molecular characteristics and causes for the low-sulfur hair disorders.

TRICHOThIODYSTROPHY SYNDROMES: DEFINITION, CLASSIFICATION, AND CLINICAL FEATURES

Definition and diagnosis
The term trichothiodystrophy (TTD) was coined by Price in 1979-19801-3 based on a series of cases, including the early report by Pollitt, Jenner, and Davies4 in 1968, of a family with mental and physical retardation and “trichorrhexis nodosa” with abnormal amino acid composition of the hair. Brown et al5 in 1970 specifically described the congenital hair defect, consisting of trichoschisis, “alternating birefringence,” and low-sulfur content. The designation for this unique hair shaft disorder is derived from Greek: tricho, hair; thio, sulfur; dys, faulty; and trope, nourishment. Clinical features of patients with TTD are highly variable in expression and severity, and phenotypes range from those with an isolated hair defect to those with severe neuroectodermal findings and, rarely xeroderma pigmentosum (XP)-like changes (Fig 1).

We reviewed the clinical features of TTD extensively in this Journal in 1990 and proposed a dysmorphic
genetic, and molecular mechanisms associated with this symptom complex have evolved substantially.\textsuperscript{7-9}

TTD, the encompassing term for the sulfur-deficient, brittle hair syndromes, is a rare disorder, inherited as an autosomal recessive trait. To date only one case with possible X-linked inheritance has been reported.\textsuperscript{10} The diagnostic findings are short, unruly, and brittle hair with low-sulfur content, alternating dark and light bands of the hair shaft under polarizing microscopy, trichoschisis, and absent or defective cuticle visualized by scanning electron microscopy\textsuperscript{11} (Fig 2). Although dark and light banding of hair shafts under polarizing light microscopy is highly suggestive for TTD, this finding is not diagnostic.

Recently, Goerz et al\textsuperscript{12} reported an 8-year-old girl with the clinical features of TTD including the “tiger-tail” pattern, visualized on polarizing microscopy and severe cuticular defects detected on scanning microscopy, but the cyst(e)ine content of the hair was normal. However, a deficiency of the sulfur-containing amino acid, methionine in the hair was documented. This case emphasizes the fact that the

Fig 1. Clinical spectrum of TTD. A, TTD and psychomotor retardation in a 2-year-old girl. TTD and photosensitivity in a 9-year-old boy showing (B) distinctive facial features and (C) hair abnormality. The patient also had ichthyosis and was found to have mutations within the XPD gene (see text).

Abbreviations used:

- CPD: cyclobutane pyrimidine dimer
- CS: Cockayne’s syndrome
- GGR: global genome repair
- ICAM-1: intercellular adhesion molecule 1
- NADPH: reduced nicotinamide adenine dinucleotide phosphate
- NER: nucleotide excision repair
- NK: natural killer
- 6-4 PP: pyrimidine (6-4) pyrimidone photoproduct
- TCR: transcription coupled repair
- TTD: trichothiodystrophy
- UDS: unscheduled DNA synthesis
- XP: xeroderma pigmentosum

relationship and classification scheme within the spectrum of the ectodermal dysplasias.\textsuperscript{6} Within the past 10 years the clinical expressions of TTD have further expanded, and our understanding of the cellular,
Morphologic abnormalities of TTD are not restricted to cyst(e)ine deficiency but also methionine and possibly other amino acid deficiencies in the hair shaft that produce a similar clinical picture.

The report of Kvedar et al. supports this contention. They observed fragile hair in two untreated patients with argininosuccinic aciduria, who showed an abnormal alternating banding pattern of the hair shafts, using polarizing microscopy. The half-cystine content was only slightly lower than in normal hair. With the institution of dietary treatment, the "tiger-tail" pattern disappeared. They coined this entity "pseudo-trichothiodystrophy" in a patient with argininosuccinic aciduria. Other inherited metabolic disorders may induce a similar phenotype. Acrodermatitis enteropathica may lead to an irregular morphologic pattern observed in the hair shaft that is characterized by alternating dark and bright bands under polarizing microscopy. After 2 years of zinc supplementation the anomaly could no longer be detected. Moreover, nonheritable alterations of hair protein composition may be seen. Amino acid content of hair may vary with season and nutrition.

A simple decrease in hair sulfur content is not diagnostic for TTD, and this entity must be recognized as a complex disease in which clinical manifestations, structural alterations of the hair shaft, and biochemical abnormalities require correlation and interpretation. Untreated kwashiorkor has been found to manifest subnormal sulfur content of hair. With treatment, the sulfur content normalized. Morganti et al. found a marked decrease in cyst(e)ine and other amino acids in the hair of several patients with different types of ichthyosis, but they did not otherwise qualify for the diagnosis of TTD. Three patients with Clouston’s ectodermal dysplasia were found to have normal hair sulfur content, but the cyst(e)ine content of hydrolyzed hair was approximately 25% to 30% lower than that of control subjects. In addition, exogenously induced and acquired types of low-sulfur hair exist. Cyst(e)ine content in hair may decrease remarkably after treatment with cold-waving lotions, depilatories, bleaching solutions, and synthetic-organic dyes.

It must be emphasized that a single, isolated morphologic abnormality of the hair shaft is not sufficient to establish the diagnosis of TTD. Although trichoschisis and alternating light and dark banding by polarizing microscopy are typical findings in TTD, they may occasionally occur in patients without this disorder.

**TTD syndromes: Delineation and spectrum of expression**

In 1968, Pollitt, Jenner, and Davies reported the first cases of “trichorrhexis nodosa,” low-sulfur content of hairs combined with mental and physical retardation. Two years later, Brown et al. described a case of trichoschisis with alternating “birefringence” (ie, light-dark banding along the hair shaft visualized by polarizing microscopy) and low-sulfur content of the hair. Trichoschisis is characterized by a sharp fracture, transversely through the entire hair shaft that is especially well detected by means of the polarizing microscope. This finding is quite typical for TTD. These investigators also discovered the marked decrease in hair sulfur content of a patient with hidrotic ectodermal dysplasia, and scanning electron microscopy revealed defective cuticle. The cuticle deformity is an additional key finding in TTD. In 1971, Tay studied 3 patients with ichthyosiform erythroderma, hair shaft abnormalities, mental and somatic growth retardation. Microscopy of hairs showed the typical findings of TTD. Several observations relating Tay syn-
drome to low-sulfur content of hair followed. At birth, children present with ichthyosiform erythroderma, and they may be encased in a collodion-like membrane. After some weeks, the erythema fades. Happle et al described translucent scaling, whereas others have observed large, dark, alligator-like hyperkeratoses. Flexures may be spared, and distinction from ichthyosis vulgaris is sometimes difficult. Histologic examination of the skin may show a thin epidermis with hyperkeratosis and absence of the granular layer, but parakeratosis or a normal granular layer with spongiosis has also been described. In 1974, Jackson, Weiss, and Watson identified a pedigree of 25 persons among an Amish kindred with brittle hair, short stature, intellectual impairment, decreased fertility, and low-sulfur content of hair. The inheritance pattern was consistent with an autosomal recessive disorder.

Cantu et al reported an apparently unrelated condition with a constellation of onychotrichodysplasia and chronic neutropenia. In 1979, three similar cases were reported. An additional patient with onychotrichodysplasia, chronic neutropenia, and mild mental retardation was subsequently described; this condition was called the ONMR syndrome. Another patient with onychotrichodysplasia, neutropenia, and normal intelligence was described. We subsequently examined a patient with chronic neutropenia, mild mental retardation, and “onychotrichodysplasia.” We further characterized the disorder as sulfur-deficient, brittle hair with trichoschisis, dark and light banding by polarizing light microscopy, and absent cuticle on electron microscopy. This observation expanded the spectrum of TTD syndromes, and Camacho designated the symptom complex as “Itin syndrome”. Over these years additional cases have been described.

Key observations along yet other seemingly separate directions had been made in the 1970s. Arbisser et al in 1976 described the Sabinas syndrome, named after a town in Mexico where most of the patients resided. They were found to have brittle hair, neuroectodermal dysplasia, and a low-sulfur content in hair. Baden et al used the acronym BIDS syndrome for brittle hair, intellectual impairment, decreased fertility, and short stature in association with low-sulfur content in the hair. Their report described hairs with an “alternating birefringent pattern” when examined by polarizing microscopy. Jorizzo et al suggested the term IBIDS syndrome for ichthyosis-associated cases in combination with the low-sulfur content hair of BIDS.

Earlier studies had documented a patient with Marinesco-Sjögren syndrome with low-sulfur content of hair. In 1971 Porter reported a case of Marinesco-Sjögren syndrome with trichoschisis and abnormal “birefringence” by polarized light microscopy. Jorizzo et al suggested the term IBIDS syndrome for ichthyosis-associated cases in conjunction with the low-sulfur content hair of BIDS. In 1979, three similar cases were reported. An additional patient with onychotrichodysplasia, chronic neutropenia, and mild mental retardation was subsequently described; this condition was called the ONMR syndrome. Another patient with onychotrichodysplasia, neutropenia, and normal intelligence was described. Chapman detected osteosclerotic abnormalities in a patient with TTD and proposed the acronym SIBIDS for osteosclerosis, ichthyosis, brittle hair, impaired intelligence, decreased fertility, and short stature. Several patients with central osteosclerosis have subsequently been described.

### Table 1. TTD subtype classification

<table>
<thead>
<tr>
<th>Type</th>
<th>Findings</th>
<th>Eponym/Acronym</th>
<th>OMIM</th>
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<tbody>
<tr>
<td>A</td>
<td>Hair +/- nails</td>
<td>Sabinas</td>
<td>211390</td>
</tr>
<tr>
<td>B</td>
<td>Hair +/- nails + mental retardation</td>
<td>Pollitt</td>
<td>275550</td>
</tr>
<tr>
<td>C</td>
<td>Hair +/- nails + mental retardation + folliculitis + retarded bone age +/- caries</td>
<td>BIDS</td>
<td>234050</td>
</tr>
<tr>
<td>D</td>
<td>Brittle hair +/- nails + infertility + developmental delay + short stature</td>
<td>Tay + BIDS</td>
<td>242170</td>
</tr>
<tr>
<td>E</td>
<td>Ichthyosis + BIDS. Hair +/- nails + mental retardation + short stature +/- decreased gonadal function +/- lenticular opacities/cataracts + failure to thrive/&quot;progeria&quot; + microcephaly +/- ataxia +/- calcifications of the basal ganglia + erythroderma and scale</td>
<td>PIBIDS</td>
<td>278730</td>
</tr>
<tr>
<td>F</td>
<td>Photosensitivity + IBIDS</td>
<td>Itin</td>
<td>258360</td>
</tr>
<tr>
<td>G</td>
<td>TTD with immune defects. Hair +/- mental retardation + chronic neutropenia or immunoglobulin deficiency</td>
<td>Itin</td>
<td>258360</td>
</tr>
<tr>
<td>H</td>
<td>Trichothiodystrophy with severe intrauterine growth retardation (IUGR). Hair + severe IUGR and failure to thrive + developmental delay + recurrent infections + cataracts + hepatic angioendotheliomatosus</td>
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Itin, Sarasin, and Pittelkow
JUNE 2001
Photosensitive trichothiodystrophy, linkage to xeroderma pigmentosum, and a unifying classification

Systematic examination of brittle hair that was detected in patients with various other phenotypic abnormalities further expanded the spectrum of TTD syndromes and eventually provided a link to another distinct group of inherited dermatologic disorders with photosensitivity and variable neurocutaneous abnormalities.

In 1983 Crovato, Borrone, and Rebora\(^67\) reported a low-sulfur hair syndrome with photosensitivity and IBIDS and suggested the acronym PIBIDS syndrome, although photosensitivity in patients with TTD had been reported earlier or concurrently.\(^68\) Another genodermatosis, xeroderma pigmentosum (XP), had been well characterized as an autosomal recessive photosensitivity disorder in which exaggerated sun damage and a very high rate of both nonmelanoma skin cancer and melanoma were characteristic features. XP was known to be associated with defective DNA repair.

Lehmann\(^69\) suggested a two-mutation hypothesis to explain the finding of defective DNA repair in TTD and to extend and relate the condition to XP. Nuzzo et al\(^70\) identified a common ancestor among families of patients with TTD and XP and they speculated that two mutations were responsible for the co-occurrence of TTD and XP type D (see later “Cellular and Molecular Genetic Characteristics of TTD: Genetic Classification of TTD” [page 900]). They concluded that if two mutations were responsible for the two diseases they are linked loci or affect the same gene.

Van Neste, Miller, and Bohnert\(^71,72\) have proposed a classification using letters alphabetically to denote the main symptoms and to separate each distinct subgroup. This classification has been updated based on recent additional findings reported in the literature and summarized in this review (Table I). This classification underscores the range as well as the overlapping consistency of features within the TTD syndromes.

Together, the TTD syndromes represent disorders within the larger spectrum of ectodermal dysplasias. Ectodermal dysplasias are a large group of heritable conditions characterized by a congenital dysplasia of one or more ectodermal structures and their accessory appendages. Although protean in expression, distinct combinations of abnormalities are observed in TTD syndromes and demonstrate consistency within the families that have been reported. The ectodermal dysplasias, as a rule, are not pure “one-layer diseases.” Mesodermal and rarely endodermal dysplasias coexist. Embryogenesis exhibits distinct tissue organiza-

Clinical manifestations and new findings in TTD

TTD is a classic differential diagnosis in congenital alopecias.\(^73\) The hair of patients with TTD is dry and sparse and the hair shafts break easily with trauma.\(^6\) Other environmental factors and mechanical stress also play roles. Intermittent hair loss during infections was observed by Kleijer, Beemer, and Boom\(^76\) and Foulc et al.\(^77\) In addition, hair loss may occur with periodic cyclicity, especially in patients with a concomitant DNA repair defect.\(^74\) Fractures of the hair shaft develop, and the viscoelastic parameters of hair are compromised compared with the hair of controls.\(^78,79\)

No effective treatment has been found for the brittle hair. In this regard Przedborski et al\(^80\) attempted treatment with oral biotin (0.75 mg/kg per day) without improvement. However, trauma and mechanical/environmental stresses should be minimized. Future treatment options include application of agents or enzymes that induce chemical modification and cross-linking of hair proteins to bridge fragile sites within the shaft and protect the cuticle from excessive damage and breakdown.

Forty percent to 50% of patients with TTD exhibit marked photosensitivity. Photosensitive patients with TTD have a deficiency in DNA excision repair, which, in most cases, is indistinguishable from that observed in XP type D.\(^77,81-86\) There is no evidence for exaggerated development of skin cancers in patients with TTD, whereas there is for patients with XP type D. Severe neuroectodermal disorders frequently occur in photosensitive TTD, but none is a constant feature. Sulfur-deficient, brittle hair remains the key finding and the objective marker for a broadening range of associated autosomal recessive ectodermal and neuroectodermal diseases, although isolated cases of TTD without other defects have been reported in recent years.\(^87-89\)

The clinical spectrum of associated signs and symptoms that constitute the TTD syndromes is extensive (Table II). Since our last review in 1990, the following additional clinical associations have been observed: expansion of neurologic abnormalities, including autism\(^41\) and partial agenesis of corpus callosum.\(^90\) Central nervous system dysmyelination occurs quite commonly, and this feature bears a similarity to Cockayne syndrome (CS).\(^52,54,55,52,64,65,91-94\) Wetzburger et al\(^56\) documented gray matter heterotopia and acute necrotizing encephalopathy in a 3-
Table II. Trichothiodystrophy syndromes: Associated signs and symptoms

<table>
<thead>
<tr>
<th>Hair*</th>
<th>Nervous system (cont’d)</th>
<th>Nervous system (cont’d)</th>
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<tr>
<td>Sparse or absent eyelashes, eyebrows</td>
<td>Lethargy</td>
<td>Perimedullary fibrosis of spinal cord</td>
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<tr>
<td>Sparse or absent axillary, pubic, body hair</td>
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<tr>
<td>Few vibrissae and otic hair</td>
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<thead>
<tr>
<th>Nails†</th>
<th>Dysmorphology and miscellaneous abnormalities‡</th>
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<tbody>
<tr>
<td>Dysplasia (onychodystrophy)</td>
<td>Cranial dysplasia</td>
</tr>
<tr>
<td>Splitting (onychoschizia)</td>
<td>Microdolichocephaly/microcephaly</td>
</tr>
<tr>
<td>Koilonychia</td>
<td>Protruding ears</td>
</tr>
<tr>
<td>Ridging</td>
<td>Hypoplastic ears</td>
</tr>
<tr>
<td>Thickening (onychogryphosis)</td>
<td>Preauricular pits</td>
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<tr>
<td>Yellow discoloration</td>
<td>Cleft ear lobes</td>
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<tr>
<td>Unguis inflexus</td>
<td>Ear deformation not specified</td>
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<table>
<thead>
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<th>Cutaneous‡</th>
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<tr>
<td>Ichthyosis</td>
<td>Growth retardation</td>
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<td>Follicular keratosis</td>
<td>Cranial dysplasia</td>
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<td>Collodion baby</td>
<td>Microdolichocephaly/microcephaly</td>
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<tr>
<td>Erythroderma</td>
<td>Protruding ears</td>
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<td>Photosensitivity (defective DNA repair)</td>
<td>Hypoplastic ears</td>
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<td>Erythema</td>
<td>Preauricular pits</td>
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<td>Eczema</td>
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<td>Hypohidrosis</td>
<td>Cleft ear lobes</td>
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<td>Pruritus</td>
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<tr>
<td>Freckles</td>
<td>Thinning of the nose</td>
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<td>Telangiectasia</td>
<td>Receding chin</td>
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<td>Hemangioma</td>
<td>Maxillary hypoplasia</td>
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<td>Lipoatrophy</td>
<td>Dental abnormalities</td>
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<td>Parchment-like skin</td>
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<td>Poikiloderma</td>
<td>Enamel hypoplasia</td>
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<tr>
<td>Folliculitis</td>
<td>Gastrointestinal malabsorption by jejunal atrophy</td>
</tr>
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<td>Chelitis</td>
<td>White plaques on tongue</td>
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<td>Hyperpigmented eyelids</td>
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<td>Hypopigmented macules</td>
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<td>Pyoderma</td>
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<td>Palmar pustules</td>
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<td>Ataxia</td>
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<td>Pyramidal signs</td>
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<tr>
<td>Muscle tone diminished</td>
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<td>Partial agenesis of corpus callosum</td>
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<td>Gray matter heterotopia and necrotizing encephalopathy</td>
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<td>Jerky eye movements</td>
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<tr>
<td>Seizures</td>
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<td>Neurosensory hearing impairment</td>
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<td>Irritability</td>
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<thead>
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<th>Ocular¶</th>
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<td>Epicanthal folds</td>
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<td>Retinal dystrophy</td>
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<td>Hypotelorism</td>
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<td>Exophthalmus/enophthalmus</td>
<td>Microdolichocephaly/microcephaly</td>
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<td>Esotropia</td>
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<td>Myopia</td>
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<td>Astigmatism</td>
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<td>Diminished red-green discrimination</td>
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<td>Strabismus</td>
<td>Growth retardation</td>
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<tr>
<td>Hypertelorism</td>
<td>Protruding ears</td>
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year-old boy with TTD. Extreme failure to thrive and death have been observed by Petrin, Meckler, and Sybert and has led to the addition of the new “H” subgroup in the classification of TTD (Table I).

Hematologic changes such as sideroblastic anemia and eosinophilia have been reported. TTD associated with right-sided hydronephrosis, ureteral duplication and left pyelocaliceal ectasia, as well as primary hypercalciuria is an additional clinical subset described recently. Gastrointestinal malabsorption with atrophic villi noted on jejunal biopsy findings and multiple food intolerance without celiac disease, requiring prolonged parenteral and enteral nutrition, has recently been observed. Abnormal results of gonadal function tests in response to luteinizing hormone-releasing hormone have been documented in patients with TTD by Przedborski et al. A practical clinical observation by O’Brien and Wilhelmus highlights ophthalmologic intervention to prevent bacterial keratitis induced by brittle, mis-oriented eyelashes.

Angioendotheliomas of the liver have been incorporated into the list of associated systemic developmental abnormalities recently described in TTD. Cleft lip, meibomian gland inflammation, and blepharitis as well as poikiloderma and progeroid facies have also been observed. Bodemer et al recently described a patient with mitochondrial disease and TTD. Initially, collodion baby was believed to occur only in patients with TTD without XPD mutation, but Marinoni et al described a case of TTD with collodion baby and mutation in the XP group D.

<table>
<thead>
<tr>
<th>Table II. Cont’d</th>
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<td><strong>Ocular (cont’d)</strong></td>
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<td>Bacterial keratitis</td>
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</tr>
<tr>
<td>Pale optic disc</td>
</tr>
<tr>
<td>Microcornea</td>
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<tr>
<td><strong>Pulmonary</strong></td>
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<tr>
<td>Pulmonary adenomatosis</td>
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<tr>
<td>Asthma</td>
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<tr>
<td>Bronchiectasis</td>
</tr>
<tr>
<td><strong>Skeletal</strong></td>
</tr>
<tr>
<td>Genu valgum</td>
</tr>
<tr>
<td>Coxa valga</td>
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<tr>
<td>Valgus deformity of the great toe</td>
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<tr>
<td>Pes valgus</td>
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<tr>
<td>Cubital and tibial valgus deformity</td>
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<tr>
<td>Ulnar deviation of fingers</td>
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*References 1, 5, 32, 42, 47, 48, 52, 64, 66, 80, 97, 99, 105, 204-208.
†References 1, 4, 32, 33, 36, 38-40, 42-44, 46-50, 52, 57, 77, 96, 97, 130, 203, 204, 208-214.
‡References 1, 4, 10, 30, 32-36, 38-44, 47, 49, 50, 52, 53, 57, 63, 65-68, 76, 77, 84, 86, 90-92, 96, 97, 100, 102, 105, 106, 109, 120, 130, 136, 137, 185, 206-210, 212, 214-237.
| **References 33, 38-41, 43-45, 63, 97, 130, 204, 207, 212, 214, 219, 239.**
| **References 1, 40, 77, 209, 215, 216.**
| **References 32, 36, 39, 42, 43, 48, 53, 80, 96, 102, 207, 209, 212, 218, 225.**
| **References 1, 4, 46, 66, 102, 206, 211, 212.**
| **References 5, 30, 38, 46-50, 63, 95-97, 99, 185, 194, 205, 209, 211, 216, 225, 240.**
Within the past few years, it has been shown that "tiger-tail" pattern on polarized hair microscopic examination also may be found in healthy infants, and therefore amino acid analysis that quantitates sulfur, specifically cyst(e)ine levels, remains the definitive test for TTD("10,11) (Table III). In this regard Garcia-Hernandez and Moreno-Giménez(112) have documented alternating dark and white zones within the hair shaft of a young patient who scratched his scalp intensely. Cessation of scratching, topical application of minoxidil 2% solution, and cysteine supplementation resulted in marked improvement within a year. The condition appears sufficiently different by polarizing light microscopy, and this sulfur-deficient hair alteration is referred to as "pseudo tiger-tailing."

The structural abnormality that causes the interrupted transverse bright lines along the hair shaft is not completely understood. However, Calvieri et al(113) and Rossi et al(114) found by x-ray microanalysis an alternating content of sulfur along the long axis of the trichothiodystrophic hair. Image analysis was also used to match the same regions that were examined by polarized microscopy and scanning electron microscopy. The x-ray analysis results also showed that calcium was absent in tracts corresponding to dark bands, whereas it was normally present in light bands.115 Definitive confirmation of these findings is awaited.

Scanning electron microscopy also shows incomplete or absent cuticle and longitudinal grooving.6 Transmission electron microscopy from hairs of patients with TTD shows material that resists extraction throughout the cortex.116 Cross-sectional examination of the cuticle in hair shows lack of the exocuticle and A layer. Transmission electron microscopy demonstrates an abnormal arrangement of microfibrils.65 Absence of the exocuticle and the sulfur-rich, A layer (outer aspect of the cuticle cell) causes cuticular weathering and weakness of the hair shaft117-119 (Table III).

Table III. Hair analysis in TTD

<table>
<thead>
<tr>
<th>Microscopy</th>
<th>Sulfur analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Light: Trichoschisis, trichorrhexis nodosa</td>
<td>Semiquantitative: Scanning electron microscopy with electron-probe microanalysis</td>
</tr>
<tr>
<td>Polarizing: Light and dark banding—tiger-tail pattern, trichoschisis</td>
<td>Quantitative: Amino acid analysis of hydrolyzed hair</td>
</tr>
<tr>
<td>Scanning electron: Transverse fracture through the hair shaft, poor or absent cuticle</td>
<td></td>
</tr>
</tbody>
</table>

Poor prognosis in TTD has been linked to severe, recurrent infectious disease with most pediatric deaths due to overwhelming bacterial infections.57

HAIR AND SKIN ABNORMALITIES IN TTD

New findings in light microscopy and scanning electron microscopy of hair

In patients with TTD, hair abnormalities are the only obligatory and diagnostic findings that identify the sulfur-deficient neuroectodermal dysplasias. Scalp hairs, eyebrows, and eyelashes are brittle, unruly, of variable lengths, easily broken, and generally feel dry. It is important to investigate the proximal parts of hair shafts because the distal portions often show marked weathering that may produce findings similar to TTD.105 Macroscopic alterations are observed especially in the frontal and occipital hair, with only microscopic abnormalities detected in the occipital hair.41 For adequate diagnosis, hairs should be collected from different areas of the scalp and subjected to further light and electron microscopic examination27,104 (Table III).

Light microscopy reveals clean transverse fractures through the hair shafts (trichoschisis), and there is an irregular hair surface and diameter.68 In addition, a decreased cuticular layer with twisting and a nodal appearance may mimic trichorrhexis nodosa.1,64,71,103-105 The distal hair shaft often terminates in "brush breaks. "103 The flattened hair shafts tend to fold over like a ribbon or shoelace during microscopic mounting. An abrupt 180° twist of the hair shaft is sometimes observed, mimicking pili torti. Polarizing microscopy with crossed polarizers shows the typical appearance of alternating light and dark bands, giving a "zig-zag" or "tiger-tail" pattern.5,6,43,106-108 The term alternating birefringence incorrectly describes the phenomenon and has therefore been abandoned. Brusasco and Restano109 reported the interesting finding that the typical “tiger-tail” pattern of the hair shaft in TTD may not be present at birth. This classical pattern was clearly evident only at 3 months of age in their case. However, hair examination from a 21-week gestation, aborted fetus showed the alternating light and dark banding pattern under polarized light microscopy.105 Within the past few years, it has been shown that "tiger-tail" pattern on polarized hair microscopic examination also may be found in healthy infants, and therefore amino acid analysis that quantitates sulfur, specifically cyst(e)ine levels, remains the definitive test for TTD("10,11) (Table III). In this regard Garcia-Hernandez and Moreno-Giménez(112) have documented alternating dark and white zones within the hair shaft of a young patient who scratched his scalp intensely. Cessation of scratching, topical application of minoxidil 2% solution, and cysteine supplementation resulted in marked improvement within a year. The condition appears sufficiently different by polarizing light microscopy, and this sulfur-deficient hair alteration is referred to as "pseudo tiger-tailing."

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Transmission electron microscopy and gene alterations of skin

Only a few studies on the ultrastructural aspects of the skin in TTD have been undertaken.42,120
These observations focused on a peculiar feature of ichthyotic skin in patients with TTD. In both patients, notable findings were similar and showed perinuclear vacuoles within unit membranes of keratinocytes and dispersed, irregularly arranged bundles of tonofilaments, particularly at the desmosome junction. The authors concluded that the abnormalities of tonofilaments could be explained by the generalized abnormality in sulfur-containing proteins, including disruption in the synthesis of keratins. Analysis of gene expression in the TTD mouse model (see “Transgenic and Knockout Mice,” page 910) has demonstrated that at least the cutaneous changes, such as acanthosis and hyperkeratosis, are associated with reduced transcription of the skin-specific, differentiation-related gene SPRR2, a member of the small proline-rich protein (SPRR) family expressed in epidermis.121 The SPRR2 gene encodes a structural component of the cornified envelope and is expressed in the final stage of terminal differentiation.122,125 Reduced SPRR2 expression in TTD skin reflects defective gene transcription in late stages of terminally differentiating epidermal keratinocytes.124

**Biochemical changes of hair shaft**

The mammalian hair follicle develops embryologically from the surface ectoderm and epidermis.125 The hair shaft is composed anatomically of the cuticle, cortex, and medulla. Hair is composed of two major structural protein families contained within the cortex predominantly, the keratins and keratin-associated proteins (KAPs), which are further classified into multiple (at least 11) subfamilies126 (Table IV). The keratin intermediate filament proteins and KAPs form the cuticle and cortex of the hair shaft.127 Keratin intermediate filaments belong to the superfAMILY of proteins that form 8- to 10-nm filaments in the cytoplasm of many epithelial cell types. Based on the amino acid composition, keratin intermediate filament proteins are classified as acidic or basic-neutral types.126 KAPs are divided into two groups, cyst(e)ine-rich and glycine-tyrosine–rich polypeptides, according to the amino acid composition of these proteins. Cyst(e)ine-rich KAPs contain high-sulfur (15%-30%) proteins (KAP1-3 or HSp), and ultra-high-sulfur proteins (KAPs 4 and 5 or UHSp) are composed of more than 30% cyst(e)ine residues.128 Other members of the KAP family as well as trichohyalin and other proteins of the hair shaft are continuing to be identified and characterized. Powell and Rogers126 have proposed a comprehensive classification scheme for hair proteins.

Total hair sulfur values were reported as early as 1806 by Vauquelin. Kutner, Miller, and Brown29 performed a systematic analysis of hair sulfur content in numerous hair shaft disorders, and they compared the results with many controls. To date, TTD is the only disease entity in which a marked decrease of sulfur content of hair composes part of the diagnostic criteria.

Amino acid analysis of hair shows a cyst(e)ine and high-sulfur protein content that is much lower than normal. In particular, the cyst(e)ine-rich proteins of TTD have lost the large heterogeneous KAP4-5 or UHSp group and at least 8 major KAP 1-3/HSp components.128 As a rule, hair of patients with TTD shows at least a 50% decrease in cyst(e)ine and sulfur content. Often, more marked decrease to less than 10% of the normal value is found. Urine and serum levels of these amino acid constituents usually are normal, but the nails may also show a decrease in cyst(e)ine and sulfur content.1 Frequently, serine, threonine, and proline are also reduced; these amino acids are components of the KAP 1,2,3.41,44,95 A concomitant, related increase in aspartic acid, methionine, phenylalanine, alanine, leucine, and lysine may be found.129 The low-sulfur protein components of hair in patients with TTD appear to be almost identical to that of normal control subjects.

<table>
<thead>
<tr>
<th><strong>Table IV. Constituent proteins of the hair shaft</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Keratin or intermediate filament (KRT or IF)</strong></td>
</tr>
<tr>
<td>KRT 1.1-1.9, type I-(acidic)—human hair “acidic”(hHa) types</td>
</tr>
<tr>
<td>KRT 2.9-2.17, type II-(basic)—human hair “basic-neutral” (hHb) types</td>
</tr>
<tr>
<td><strong>Keratin- or IF-associated proteins (KRTAP/KAP or IFAPs)</strong></td>
</tr>
<tr>
<td>Cysteine-rich group (cys-KAPs)</td>
</tr>
<tr>
<td>High sulfur</td>
</tr>
<tr>
<td>KAP 1 (B2) family</td>
</tr>
<tr>
<td>KAP 2 (BIIIA) family</td>
</tr>
<tr>
<td>KAP 3 (BIIIB) family</td>
</tr>
<tr>
<td>Ultra-high sulfur</td>
</tr>
<tr>
<td>KAP 4 family</td>
</tr>
<tr>
<td>KAP 5 family</td>
</tr>
<tr>
<td><strong>Cuticle</strong></td>
</tr>
<tr>
<td>KAP 10 family</td>
</tr>
<tr>
<td>Glycine/tyrosine-rich group (gly/tyr-KAPs)</td>
</tr>
<tr>
<td>KAP 6 (type II) family</td>
</tr>
<tr>
<td>KAP 7 (type I C2) family</td>
</tr>
<tr>
<td>KAP 8 (type I F) family</td>
</tr>
<tr>
<td><strong>Other proteins</strong></td>
</tr>
<tr>
<td>KAP 9 (mouse ultra-high sulfur) family</td>
</tr>
<tr>
<td>KAP 11.1 (novel mouse hair protein)</td>
</tr>
<tr>
<td><strong>Medulla proteins</strong></td>
</tr>
<tr>
<td>Trichohyalin</td>
</tr>
<tr>
<td>Calcyclin</td>
</tr>
<tr>
<td>Involucrin</td>
</tr>
</tbody>
</table>

Modified from Powell and Rogers126 and Bertolino and O’Guin.243
but occasionally they are higher than normal.\textsuperscript{130} The KAP 1-3/HSps are altered qualitatively, and, moreover, the KAP 4-5/UHSps are severely decreased.\textsuperscript{131} One-dimensional electrophoresis analysis by sodium dodecyl sulfate–polyacrylamide gel electrophoresis followed by fluorography shows that levels of high-molecular-weight basic-neutral keratins are generally preserved among hair diseases when compared with their acidic keratin partners and to KAP 1-3/HSps and KAP 4-5/UHSps.

Hairs in TTD are characterized by loss of the large heterogeneous KAP 4-5/UHSps group including the 33- and 42-kd proteins and at least 8 major KAP 1-3/HSps components. The expression of the 54-kd type 1 keratin and the 38-kd KAP 1-3/HSps varies among patients with TTD.\textsuperscript{132} These findings provide evidence for the concept of heterogeneity of TTD. Gillespie, Marshall, and Rogers\textsuperscript{133} observed in their analysis of hair by two-dimensional protein electrophoresis that the pattern of KAP 1-3/HSps and KAP 4-5/UHSps in TTD differed from normal controls but also among individual patients.

The typical brittleness of TTD hair likely results from a reduction in the content of the hair-specific cyst(e)ine-rich proteins, KAP 1-3/HSps, and KAP 4-5/UHSps that fill the spaces within the matrix. The assembling keratin filaments form the microfibrils, and the high proportion of cyst(e)ine residues, ranging from 15 to 70 or more, in the KAPs likely promote formation of multiple covalent disulfide bonds within and between these proteins. De Berker, Tolmie, and Dawber\textsuperscript{110} concluded that the intrinsic defect is due to failure of incorporation of sulfur-rich protein into the cuticle and matrix of the hair cortex. KAP 1-3/HSps and KAP 4-5/UHSps can be modified qualitatively or quantitatively as in a TTD variant defined by Van Neste et al.\textsuperscript{130} This contrasts with the qualitative and quantitative alterations observed in classic TTD. Van Neste et al.\textsuperscript{129} and De Brouwer et al.\textsuperscript{114} showed that the amino acid composition of hairs collected from a patient with a TTD variant was preserved when follicles had been grafted and maintained up to 6 months on nude mice. The persistence of disease-specific abnormalities within the hair shaft indicates that the TTD-variant mutation is constitutively expressed within the hair follicle unit and is independent of host-related factors.

Deficiency in cyst(e)ine residues and decrease in the fraction of KAP 4-5/UHSps of the matrix are consistent abnormalities within the hair shaft of TTD. In addition to the abnormal low-sulfur distribution in the cortex, sulfur deficiency has also been localized to the cuticle cells. In cuticle cells from TTD hair, the exocuticle appears less dense and the A layer is absent or greatly reduced in thickness.\textsuperscript{116} These changes seem to reduce the mechanical strength of the cuticle cells. Cuticle is lost, and the cortex becomes vulnerable to weathering. Hair color, however, appears to be unaffected. In the future it would be worthwhile to analyze sulfur content in the hair of patients with XP and CS, although clinically they do not feature marked brittleness of hair shafts.

**Prenatal Diagnosis**

Selected types of TTD manifest significantly more severe and potentially lethal phenotypes. In these cases, prenatal diagnosis and therapeutic abortion or other interventions have been considered. Approximately 50% of patients with TTD show photosensitivity and reduced DNA repair levels similar to those found in XP.\textsuperscript{135} Under these circumstances, prenatal diagnosis based on measurement of DNA repair in trophoblasts or amniotic cells and subsequent confirmation by microscopic analysis of fetal hairs has been performed.\textsuperscript{76,105,136,137} Examination of hair by polarized light microscopy of an aborted fetus demonstrated the alternating light and dark bands typically seen in TTD.\textsuperscript{105} The conventional procedure to assay unscheduled DNA synthesis requires 4 to 5 weeks and is labor intensive. Alapetite et al.\textsuperscript{138} recently proposed the more rapid, yet sensitive comet assay as a DNA repair test for prenatal diagnosis of TTD. This examination can be performed as a single-cell gel electrophoresis assay that typically provides a result within 24 hours and avoids radioactive substances.

**Cellular and Molecular Genetic Characteristics of TTD**

On treatment with DNA-damaging agents, it is possible to detect and further characterize cellular abnormalities linked to nucleotide excision repair (NER) defects with the use of end points such as reduced levels of DNA repair synthesis, decreased cell survival, decreased rates of DNA and RNA synthesis, and increased mutability.\textsuperscript{139} Cells from patients with NER defects are usually assigned to a designated complementation group by means of the somatic cell fusion assay that measures the level of unscheduled (separate from the S phase of the cell cycle where DNA replication occurs) DNA synthesis (UDS) after UV irradiation of fused heterokaryons. Cell fusion studies or expressed, cloned excision repair genes after microinjection\textsuperscript{140,141} or retroviral infection\textsuperscript{142} have disclosed genetic heterogeneity among patients with defective NER. Although there is considerable molecular-genetic overlap among DNA repair–deficient patients (Fig 3), clinical features may differ dramatically.\textsuperscript{143} A major discovery in advancing the understanding of the genetics of TTD...
Fig 3. Clinical and genetic heterogeneity in patients with XP, CS, and TTD. The genetic heterogeneity between the 3 diseases is shown by the overlap between XP and XP/CS due to mutation on the \( XPB \), \( XPD \), or \( XPG \) genes and between XP and TDD due to mutation on the \( XPB \) or \( XPD \) genes.
Fig 4. Model of the global genomic repair (GGR, left part) and transcription-coupled repair (TCR, right part) of UV-induced DNA lesions. The complex XP-C/HR23B is the first factor in GGR only to bind DNA lesions and to attract XPA, RPA (ssDNA binding protein) and then TFIIH. The XPE protein seems to facilitate the identification of lesions, which are poorly recognized by the XPC/H23R complex (such as the CPD). Demarcation of the lesions is carried out by the two helicase activities (XPB and XPD) of TFIIH followed by sequential cleavage by the two structure-specific nucleases XPG (on 3' side) and ERCC1-XPF (on 5' side). After removal of an oligonucleotide (27-30 nucleotides long) containing the lesion, DNA synthesis occurs using either polymerase δ or ε in presence of PCNA and RF-C complexes as processivity factors. The final NER step is ligation of the newly synthesized DNA patch to parental DNA probably by DNA ligase I. On damaged templates, RNA polymerase II is blocked by bulky lesions inducing a signal for TCR (right part). The proteins CSA, CSB, and possibly XPG and TFIIH displace the stalled RNA pol II from the lesion, which becomes accessible for further repair in the same way as for GGR.
syndromes was the demonstration that fibroblasts of some patients with TTD were genetically similar to those isolated from patients with XP belonging to the D group. Because patients with XP were known to be deficient in DNA repair, it was then hypothesized that the cells from patients with TTD were also DNA repair deficient.

**General aspect of the NER pathway**

NER is critical in all organisms to protect the genome against injury by numerous mutagenic and carcinogenic agents. A complex-overlapping network of enzymatic pathways for DNA repair has evolved to minimize genetic instability and initiation of carcinogenesis. Approximately 30 gene products are involved in this intricate protective mechanism. This NER system eliminates structural lesions that range from UV-induced photoproducts to chemical-produced adducts and intrastrand crosslinks. A series of steps is involved in the NER complex including recognition of the DNA lesion, removal of the damaged oligonucleotide, gap filling by DNA synthesis, and ligation (Fig 4). In addition, NER mechanisms can also recognize small oxidative adducts, as shown by Le Page et al and Leadon, that may be involved in the progressive neurologic deterioration of some patients (Tables V and VI).

**The link between DNA repair and transcription**

There is growing evidence that damage produced by UV is repaired more rapidly in transcriptionally active DNA than in the genome as a whole. This preferential repair mechanism has been shown to be due to accelerated repair of damage in the transcribed strand versus the nontranscribed strand of DNA because of the blockage of the RNA polymerase II at the site of the lesion, giving rise to a signal for rapid removal. Indeed, for a subset of DNA lesions, two overlapping pathways have been identified in the NER process. One pathway is the more rapid transcription-coupled repair (TCR) of expressed genes, targeted to the transcribed strand of DNA, and the other is the slower, global genome repair (GGR) of DNA, which includes repair of the nontranscribed strand of potentially expressed genes, as well as the inactive chromatin (Fig 4).

The predominant types of UV-induced lesions are cyclobutane pyrimidine dimers (CPD) and pyrimidine (6-4) pyrimidone photoproducts (6-4 PP), both removed by NER. In all classic XP cells, repair of both lesions is defective in all parts of the genome, except the XP group C cells that are fully proficient in the TCR of these adducts. The XPC protein is therefore not necessary for TCR because the signal for the presence of lesions is probably given by the stalled polymerase. By contrast, cells from patients with CS are able to remove these lesions in all parts of the genome except those located on transcribed strands. The CSA and CSB proteins, which are mutated in most patients with CS, are thought to allow the removal of the stalled polymerase in such a way that the lesion becomes accessible to repair enzymes. Eveno et al have shown that repair of CPD was significantly reduced in all TTD cell lines (at a similar level as in XP cells), whereas almost normal repair of 6-4 PP was found in most TTD lines except for some specific patients in whom the efficiency was

### Table V. Laboratory comparison of XP, CS, and TTD complementation groups

<table>
<thead>
<tr>
<th>Complementation group</th>
<th>UV sensitivity</th>
<th>Residual UDS</th>
<th>TCR</th>
<th>GGR</th>
<th>Overlap with other nucleotide excision syndromes</th>
</tr>
</thead>
<tbody>
<tr>
<td>TTD-A</td>
<td>+</td>
<td>15%</td>
<td>+(?)†</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>XP-A</td>
<td>++</td>
<td>&lt;5%</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>XP-B</td>
<td>++</td>
<td>&lt;10%</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>XP-C</td>
<td>+</td>
<td>15%-30%</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>XP-D</td>
<td>++</td>
<td>15%-50%</td>
<td>+</td>
<td>+</td>
<td>CS,‡ PIBIDS</td>
</tr>
<tr>
<td>XP-E</td>
<td>+/-</td>
<td>&gt;50%</td>
<td>?</td>
<td>?</td>
<td>-</td>
</tr>
<tr>
<td>XP-F</td>
<td>+</td>
<td>15%-30%</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>XP-G</td>
<td>++</td>
<td>&lt;10%</td>
<td>+</td>
<td>+</td>
<td>CS‡</td>
</tr>
<tr>
<td>CS-A</td>
<td>+</td>
<td>WT</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>CS-B</td>
<td>+</td>
<td>WT</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

Adapted from Hoeijmakers JHJ. Eur J Cancer 1994;30A:1913.

*WT*, Wild type (control level).

†XP/CS complex.

‡Decreased intracellular TFIIH.

![Table V](https://example.com/table-v.png)
The TFIIH complex regulates transcription and the cell cycle under basal conditions, but also coordinates TCR, DNA repair, or apoptosis when DNA damage is induced by UV.\(^1\) The XPB helicase is absolutely required for transcription initiation, whereas the XPD helicase is dispensable but stimulates transcription by helping XPB in promoter opening.\(^2\) Therefore, TFIIH is thought to unwind the DNA to allow promoter clearance at the site of transcription initiation and at the damage site during NER. The binding of wild-type p53 protein to XPD or XBP proteins inhibits the helicase activities of purified TFIIH in vitro.\(^3\) The same gene

---

**Fig 5.** Diagram of TFIIH complex as it inhibits cell cycle progression and transcription but induces TCR, DNA repair, or apoptosis occurring after genome damage. TTD, CS, or XP are the consequence of TFIIH malfunction. (Adapted from Moustacchi E, coordinator. DNA repair. Biochimie 1999;81:1-181.)

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...less than normal (see “Cellular responses in TTD,” page 908).

The relationship between DNA repair and transcription became obvious when Schaeffer et al.\(^4\) studied the protein structure of one of the major transcription factors, TFIIH. TFIIH is a large complex of 9 proteins involved in the last step of the initiation of transcription, as well as in the regulation of the cell cycle through its cyclin kinase activity and in the NER pathway because of the presence of the XPB and XPD proteins with helicase activities. The multiprotein complex is composed of subunits ranging in size from 32- to 89-kd (Fig 5). Two main components, the core-TFIIH subcomplex containing the XPB-p89 subunit and the subcomplex containing the kinase activity (cdk7, MAT1, and cyclin H) are associated with the XPD (80 kd) subunit. The TFIIH complex regulates transcription and the cell cycle under basal conditions, but also coordinates TCR, DNA repair, or apoptosis when DNA damage is induced by UV.

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defect can result in apparently identical cellular phenotypes related to DNA repair deficiency, yet gives rise to completely different clinical features. Mutation in one of the two DNA repair genes in TFIIH may lead to 3 different human disorders: the skin cancer–prone syndrome, XP, and TTD- or CS-associated or not with XP in the same patient (Fig 3). In addition to sun sensitivity (for a fraction of patients with TTD or CS), all these syndromes include neurologic abnormalities associated with nerve dysmyelination and severe growth retardation. TTD and CS could well be part of an enlarging group of heterogeneous disorders classified as transcription diseases (see “TTD as a Transcription Disease,” page 911).

**Genetic classification of trichothiodystrophy**

Patients with TTD can be categorized in two major groups: (1) the nonphotosensitive and defect-free in excision repair of UV damage and (2) the photosensitive with NER defect. In the first group, no gene has been isolated yet, because of the lack of a screening assay to isolate the gene(s). In the second group, 3 complementation groups have been determined. The major one, representing about 95% of the photosensitive patients, is due to mutations within the XPD gene. Two patients have mutations of the XPB gene and one patient has a mutation of a yet-unknown gene, called TTD-A. Furthermore, to date not all mutations have been identified in the photosensitive TTD group.

**The XPD gene and TTD.** The DNA repair gene, ERCC2 (according to the former nomenclature), has been identified as the gene mutated in XPD. A set of rodent mutants has been valuable as a template for the isolation of human NER genes that correct DNA repair defects; one such gene is termed ERCC2 for excision repair cross-complementing, with the numeral 2 referring to the rodent complementation group. The human ERCC2 gene homologue (XPD) maps to the long arm of human chromosome 19 within a region containing several other genes involved in DNA repair and metabolism. XPD is located on 19q13.2-q13.3 and within 2 megabases of the XRCC1 locus. In the human, the XPD gene is transcribed from centromere to telomere, in the same orientation as the nearby ERCC1 gene. The XPD gene is composed of 23 exons with a genomic distance of 18.9 kb and encodes a 760 amino acid protein with adenosine triphosphate–dependent DNA 5′-3′ helicase activity. The XPD gene is highly conserved over evolution, with the order and orientation of at least 3 genes preserved within the mammalian lineage of this linkage group. After transfer by microinjection or retroviral transduction into cells from patients with XPD or with photosensitive TTD, respectively, wild-type XPD gene was found to restore normal UV sensitivity and DNA repair to cells as well as provide partial resistance to UV-induced mutagenesis.

Weeda et al proposed that the XPD helicase unwinds the DNA in the vicinity of a lesion in the opposite direction to the XPB helicase. As already described, this protein is involved in TCR, being an integral member of the basal transcription factor TFIIH complex. Two patients with specific mutations on the XPD gene also exhibit CS, reinforcing the relationship between repair and transcription deficiencies.

Nucleotide sequence analysis of the XPD complementary DNA (cDNA) from TTD cell strains revealed mutations within the coding sequence, and particularly in highly conserved regions within previously identified helicase functional domains. To date, 47 mutations have been characterized in patients with XPD or XPD/CS and 45 mutations in patients with TTD (see Fig 6, A). These mutations are mostly point mutations leading to a single amino acid change located mainly in the C-terminal part of the protein. The various clinical presentations and DNA repair characteristics of the cell strains should, in theory, correlate with the particular mutations found in the XPD locus. However, because these repair-deficient diseases are transmitted as a recessive trait, the two alleles of the same gene have to be mutated. In approximately 50% of cases, the sequenced XPD genes in patients with TTD revealed compound heterozygotes (Fig 6, A). In such cases, it is plausible that the phenotype is defined by the residual activity of the less severe of the two alleles. Therefore it was essential to determine which mutated allele was responsible for the clinical presentation. Taylor et al examined the phenotype in the haploid state using the eukaryotic yeast model Schizosaccharomyces pombe rad 15 for most of the homologues of XPD mutations found in patients with TTD and in those with XP to determine the null and the functional alleles. By extrapolation, they could deduce the human mutations directly responsible for a given phenotype and exclude the mutations that were nonviable in the yeast system (Fig 6). Analysis of Fig 6, B reveals that most causative mutations are clustered in the C-terminal fourth of the protein with the exception of a TTD hotspot close to the N-terminus. All the mutations are specific for either TTD or XP (in contrast to the mutations presented in Fig 6, A where null alleles were found in common between TTD and XP cells). Sixty percent of causative mutations for XP are located at the Arg683 hotspot, whereas 50% of TTD mutations are
Fig 6. See legend on facing page.
located at the Arg112 hotspot. The other TTD mutations are localized in the C-terminal region at 6 different, but closely linked, positions (Fig 6, B).

By reconstituting a functionally active TFIIH with recombinant polypeptides, Coin et al173 showed that the XPD helicase activity was strongly stimulated by the interaction with the p44 subunit, inside the transcription complex. This interaction occurs within the C-terminal domain of XPD and the mutations found in this part in patients with TTD or XPD do not abolish the helicase activity per se but prevent the interaction and therefore the stimulation by p44 of the helicase activity. This low helicase activity is responsible for the NER defect in these patients.173,174

The XPB gene and TTD. The ERCC3 gene has been isolated as the gene mutated in group 3 of DNA repair–deficient rodent cells. The human homologue, XPB, was able to complement the cells isolated from patients with XP group B.175 This gene is located on chromosome 2q21 and encodes 782 amino acids with a 3´-5´ helicase activity. The helicase unwinds the DNA at the lesion in parallel with the XPD protein, both present in the TFIIH complex.154,176

Only 5 patients are known to harbor mutations within this gene (Fig 7), indicating clearly that the protein is absolutely necessary for viability, as confirmed by knock-out animals which are nonviable.121,124 Any mutation that destroys the transcriptional activity of TFIIH will be lethal because, in contrast to XPD, XPB plays a crucial role in TFIIH activity. Among these patients, 3 have both XP and CS. Coin et al174 demonstrated that mutations in these patients had decreased transcriptional activity of the corresponding purified TFIIH because of promoter opening blockade. One patient, initially described in 1980 and now in his twenties, has the typical signs and symptoms of TTD, with sensitivity to sunlight since early childhood.106 His clinical features are quite distinct from those associated with XP. There is no significant freckling or other pigmen-
tary changes and no development of malignant skin tumors.143 A new NER gene associated with this TTD patient has been identified and shown to be the gene involved in XP group B.160 Another sibling showed the same defect (Fig 7).

Fig 6. Mutations in the XPD gene found in patients with TTD, XP and XP/CS. The XPD protein (761 amino acids) contains 7 domains DNA/DNA helicase (indicated by the number of amino acids involved) and 7 domains of DNA/RNA helicase (indicated in gray-shaded boxes below the protein). The mutations are indicated as the number and the type of the amino acid change. Mutations above the XPD protein correspond to XP patients (the XP/CS mutations are boxed) and below the XPD protein correspond to TTD patients. FS = frameshift and dotted line indicates deletion. The different mutated alleles in the same patient are indicated with numbers 1 and 2. A, All the XPD mutations found in the two classes of patients are indicated. B, Only causative mutations are indicated according to Taylor et al172 and Botta et al.57 The interaction domain between the XPD helicase and the p44 protein inside the TFIIH factor is indicated according to Coin et al.173,174

Table VI. Main clinical findings of NER syndromes

<table>
<thead>
<tr>
<th>Clinical symptoms</th>
<th>XP</th>
<th>XP/CS</th>
<th>CS</th>
<th>TTD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Photosensitivity</td>
<td>++</td>
<td>++</td>
<td>+*</td>
<td>+*</td>
</tr>
<tr>
<td>Skin cancer</td>
<td>++</td>
<td>+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Progressive mental degeneration</td>
<td>+/-*</td>
<td>+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neuronal loss</td>
<td>+/-*</td>
<td>+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neurodysmyelination</td>
<td>–</td>
<td>++</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thin faces</td>
<td>–</td>
<td>+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Growth defect</td>
<td>+/-*</td>
<td>+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypogonadism</td>
<td>+/-</td>
<td>+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Brittle hair and nails</td>
<td>–</td>
<td>–</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ichthyosis</td>
<td>–</td>
<td>–</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>


–, Absent; +/-, minimal; +, present; ++, marked.

*Patients with TTD and CS may have no photosensitivity and NER defect.

†Neurologic and growth defects characteristic of XP patients with DeSanctis-Cacchione syndrome.
sensitive to the cidal effects of UVC or UVB irradiation and present a reduced level of DNA repair synthesis compared with the heterozygote parent or normal cells. These two cellular characteristics allowed us to propose prenatal diagnosis of these syndromes.105

The cellular responses to UV irradiation are remarkably heterogeneous among the various TTD cell lines. Some TTD lines mutated on the \textit{XPD} or \textit{TTD-A} genes exhibit a very low survival and UDS level after irradiation, whereas other \textit{XPD}- or \textit{XPB}-mutated TTD cells presented a smaller reduction in UDS level and a better cell survival after UV.179 These variations are presumably linked to the extent of repair of CPD and 6-4 PP in these various cells.153,180

The cellular sensitivity to UV irradiation is largely dependent on the position of the functional mutation181,182 (Fig 6, A). There is no correlation between the severity of DNA repair deficiency and the clinical symptoms. The R112H hotspot, found in almost 30% of patients with TTD, is associated with a drastic reduced level of DNA repair but with moderate clinical features and no cancer sensitivity.57 Botta et al\textsuperscript{57} concluded that the severity of the clinical features might be linked to the gene dosage between the two different mutated \textit{XPD} alleles. The most severe clinical features are observed in patients with TTD who appear to be hemizygous for the mutated allele. This result implies that the transcriptional efficiency of the mutated TFIIH in TTD could be more important for the clinical features than the DNA repair level itself.

Cultured TTD fibroblasts exhibit a high mutation frequency after UV irradiation.183-185 Madzak et al\textsuperscript{183} and Marionnet et al\textsuperscript{184} documented that the fre-
frequency of mutations in TTD and XP cells are similar and therefore high levels of UV-induced mutations are not always directly related to a predisposition to cancer. In TTD cells, there are more rearrangements than in either repair-proficient or XP-D cell lines. It has been speculated that the consequences of UV-induced mutations in TTD cells could be deleterious for essential genes, resulting in cell death rather than mutation propagation and tumorigenesis.\textsuperscript{184} Outside the frequency of gene rearrangements, the types of substitutions are closer to that observed in normal cells than that of XPD cells, indicating that the mutagenesis pathway in TTD is similar to normal cells. However, the absence of repair of CPD (at least) produced a higher rate of mutations.

Otto et al\textsuperscript{186} have recently shown differential responses of primary (nontransformed) fibroblasts and keratinocytes from normal and DNA repair–deficient patients toward UVA and UVB. The same dose of UVB (1000 J/m\textsuperscript{2}) induced twice as many DNA lesions in normal fibroblasts compared with normal keratinocytes. UV survival, determined by clonal analysis, was consistently higher in keratinocytes than in fibroblasts. Normal and TTD keratinocytes survived better after UVA and UVB irradiation than keratinocyte cells from patients with XP type C or D. Furthermore, the authors showed that UV irradiation resulted in a transition from proliferative to abortive colonies. This transition, which varied between donors, could reflect a natural protection against UV-induced tumorigenesis. This process was, in part, inversely correlated to the patient’s predisposition to cancer.\textsuperscript{186}

Variable results have been reported for the efficiency of repair of specific UV-induced DNA lesions in TTD versus XP and normal cells.\textsuperscript{153,158,180,185} Basically, however, TTD cells are able to repair almost normally the 6-4 PP but are deficient in the repair of CPD, whereas the XPD cells are deficient in both repair mechanisms. The level of repair varies according to the site of mutations in the \textit{XPD} gene. For example, cells mutated at the R722W position are virtually “wild type” for the 6-4 PP repair, whereas cells homozygous at the hotspot R112H have a repair deficiency approximating the XPD cells.\textsuperscript{168} This finding explains why cells from patients with TTD have only a mild defect in survival after UV irradiation, whereas the XPD lines are very UV sensitive. CPD and 6-4 PP are repaired in a rapid and complete fashion by the TCR machinery in the transcribed strand of active genes. Elsewhere in the genome, repair by GGR is slower and less efficient.\textsuperscript{152} Dumaz et al\textsuperscript{187} have shown that accumulation of the p53 tumor suppressor gene product is markedly enhanced after UV radiation in cells of XP as well as in TTD and CS. p53 is stabilized with a much lower amount of UV in TTD, CS, and XPD cells versus normal cells and for a much longer time (3-4 days instead of 16 hours), thereby indicating that the stabilization of the p53 protein and the blockage of the cell cycle is due to the presence of DNA lesions on the transcribed strands of active genes and probably to the presence of unrepaired CPD, which inhibits RNA polymerase II progression. In addition, the same authors documented recovery of the normal p53 response after UV treatment in DNA repair–deficient fibroblasts by retrovirally mediated correction with the \textit{XPD} gene.\textsuperscript{187} These results were confirmed by a study by Abrahams et al\textsuperscript{188} showing differences in the regulation of p53 stability in UV-irradiated normal versus DNA repair–deficient human cells. The authors showed that normal fibroblasts exhibit a transient and UV dose-dependent stabilization of p53, but fibroblasts from repair-deficient syndromes show abnormalities in p53 protein stability. Although it has been shown that XPD and XPB cells are deficient in p53-induced apoptosis,\textsuperscript{158} we

### Table VII. Similarities and differences between XPD and TTD/XPD cells after UV irradiation

<table>
<thead>
<tr>
<th>Cellular response</th>
<th>XPD cells</th>
<th>TTD/XPD cells*</th>
</tr>
</thead>
<tbody>
<tr>
<td>UV survival</td>
<td>Very low</td>
<td>Low</td>
</tr>
<tr>
<td>UDS</td>
<td>20%-40%</td>
<td>15%-30%</td>
</tr>
<tr>
<td>Mutation frequency</td>
<td>High</td>
<td>High</td>
</tr>
<tr>
<td>Spectrum of base substitutions</td>
<td>Different from WT</td>
<td>WT</td>
</tr>
<tr>
<td>Repair of CPD</td>
<td>Low</td>
<td>Low</td>
</tr>
<tr>
<td>Repair of 6-4 PP</td>
<td>Low</td>
<td>WT†</td>
</tr>
<tr>
<td>p53 Responses</td>
<td>Low UV dose</td>
<td>Low UV dose</td>
</tr>
<tr>
<td>UV-apoptosis induction</td>
<td>Low UV dose</td>
<td>Medium UV dose</td>
</tr>
<tr>
<td>Catalase activity</td>
<td>Low</td>
<td>WT</td>
</tr>
<tr>
<td>Induction of ICAM-1</td>
<td>Low</td>
<td>WT†</td>
</tr>
</tbody>
</table>

\textsuperscript{WT}, Wild type.

\*For photosensitive patients.

†The TTD cells mutated at the site R112H are closer to the XPD cell responses than the other TTD cells.
found that low doses of UVC or UVB induced much more apoptosis in XPD fibroblasts than in TTD (Queille S et al, J Invest Dermatol; in press). If keratinocytes behave as fibroblasts do, this suggests that the absence of skin cancer in TTD is not due to enhanced levels of apoptosis in damaged cells. The high level of apoptosis in XPD cells irradiated with low UV doses stimulates cell-cycle turnover in the irradiated epidermis, compensating for the decrease in total cell number and leads to DNA replication in stem cells containing DNA lesions. This process is mutagenic in itself and may be carcinogenic as well, which explains the predisposition to cancer in patients with XPD.

Similarities and differences between XP type D and TTD cells after UV exposure are summarized in Table VII.

**Table VIII. Characteristics between NER mouse models and corresponding human syndromes**

<table>
<thead>
<tr>
<th>Gene</th>
<th>Mouse mutation</th>
<th>UV sensitivity</th>
<th>UDS (%)</th>
<th>Skin cancer</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Man</td>
<td>Mouse</td>
<td>Man</td>
</tr>
<tr>
<td>XPA</td>
<td>KO</td>
<td>+++</td>
<td>+++</td>
<td>&lt;5</td>
</tr>
<tr>
<td>XPB</td>
<td>KO*</td>
<td>N/A</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td>XPC</td>
<td>KO</td>
<td>+</td>
<td>+</td>
<td>15-30</td>
</tr>
<tr>
<td>XPD</td>
<td>KO*</td>
<td>N/A</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td>XPD</td>
<td>TTD point mutation</td>
<td>+/−</td>
<td>+/−</td>
<td>25</td>
</tr>
<tr>
<td>CSB</td>
<td>Truncation</td>
<td>++</td>
<td>++</td>
<td>Normal</td>
</tr>
<tr>
<td>CSA</td>
<td>KO</td>
<td>++</td>
<td>++</td>
<td>Normal</td>
</tr>
<tr>
<td>ERCC1</td>
<td>KO truncation†</td>
<td>N/A</td>
<td>+++</td>
<td>N/A</td>
</tr>
<tr>
<td>mHR23a</td>
<td>KO</td>
<td>N/A</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td>mHR23b</td>
<td>KO</td>
<td>N/A</td>
<td>−</td>
<td>Normal</td>
</tr>
</tbody>
</table>

Adapted from de Boer J, Hoeijmakers JH. Biochimie 1999;81:127-37.

KO, Knockout; N/A, not applicable; ND, not done.

*Early embryonic lethality.

†Die before weaning.

**TRANSGENIC AND KNOCKOUT MICE**

Because no animal model had been available to mimic human DNA repair–deficient diseases, transgenic mice and, subsequently, knockout animals have been produced to study the biologic consequences of repair deficiency in animals. NER-deficient mice have recently been generated, giving rise to phenotypes close to XP, CS, and TTD. As expected, XPA and XPC knockout or null animals were very close to the human phenotypes, showing UV sensitivity and predisposition to cancer, whereas XPD and XPB null animals were nonviable, resulting in very early embryonic lethality and showing clearly the indispensable role of these two helicases for normal life. A mouse model for CS, with selective impairment of TCR, has been produced. These animals mimic very well the human phenotype, except they are prone to skin cancers whereas the human counterparts are not (Table VIII).

A transgenic mutant with a partial repair defect and associated clinical symptoms similar to TTD was created by mimicking a point mutation, identified as one of the hotspots in the XPD gene, of a photosensitive patient with TTD (mutation R722W, see Fig 6, A) and using a novel gene cDNA fusion targeting strategy. TTD mice reflect, to a remarkable degree, the human disorder, including growth retardation, developmental abnormalities, reduced life span, skin abnormalities, and prematurely aged appearance. Low cysteine content in hair, hair loss, and brittle hair were similar to that found in patients with TTD. Cellular and skin UV sensitivity and low UDS level are similar to that observed with the corresponding human TTD cells. The reduced transcription of the specific SPRR2 gene, involved with the cross-linking process of the cornified envelope and expressed in the final stage of terminal differentiation, strongly supports the concept of TTD as a human disease due to inborn defects of basal gene transcription and DNA repair. However, De Boer et al recently found that, in striking contrast to human TTD, the TTD mouse is clearly susceptible to UV- and 7,12-dimethylbenz[a]anthracene–induced skin carcinogenesis, although at a lower level than the XPA or XPC mouse. These findings suggest that patients with TTD could harbor a predisposition to skin cancer, though this is not in agreement with the human clinical experience. Long-term clinical surveillance will be necessary to more clearly delineate this possible risk. However, it might well be that the human genome and/or cells have developed inherent protective mechanisms not
present in lower order species, such as the mouse. Nonetheless, these animal models provide helpful tools to understand the complex relationships between DNA repair defects, transcription function, and clinical manifestations.

**LACK OF PREDISPOSITION TO CANCER IN TTD**

It is clear that during their lifetimes, patients with TTD with DNA repair deficiency as well as those with CS do not develop excess malignancies, whereas patients with classic or variant XP with similar DNA repair defects are predisposed to numerous malignancies.\(^{166,167,179}\)

Vuillaume et al\(^ {192}\) reported striking differences in cellular catalase activity between XP and TTD. In XP fibroblasts, catalase activity was 5-fold less than that in controls. Fibroblasts of patients with TTD showed a high level of catalase activity. However, molecular analysis of catalase gene or messenger RNA accumulation showed no difference between normal, XP, and TTD cells. The low catalase activity has been shown to be due to an intrinsic low level of the intracellular concentration of reduced nicotinamide adenine dinucleotide phosphate (NADPH), for yet unknown reasons, which is an obligatory cofactor of catalase. Therefore this low level of NADPH is directly responsible for the low catalase activity and is specific for XP cells versus TTD or normal cells. Growth of XP cells in the presence of 0.1 mmol/L of NADPH fully complements the catalase deficiency in XP cells.\(^ {193}\) UV irradiation induced 3 to 5 times more intracellular hydrogen peroxide production in XP cells compared with TTD cells or controls. These striking differences were interpreted by the authors as showing that UV radiation, directly or indirectly, together with defective oxidative metabolism may increase the initiation and/or the progression steps in the XP cellular environment compared with TTD.\(^ {192}\)

Terleth et al\(^ {194}\) attempted to explain the absence of cancer predisposition in patients with TTD as well as in patients with XP by the lack of induction of the ornithine decarboxylase gene, a putative proto-oncogene, and the absence of the induction of mammalian SOS-like response, after irradiation: these two events seem to be necessary for a cell response leading to cancer. However, the molecular reason for this lack of induction is not yet understood.\(^ {195}\)

One of the major processes involved in cancer development and progression is the efficiency of host immune surveillance. It is known that immunodeficient patients are predisposed to skin cancers on sun-exposed body parts. Similarly, an impairment of cell-mediated immunity has been proposed as a cofactor in the predisposition to cancer of patients with XP. For example, Mariani et al\(^ {195}\) documented that the relative proportion of CD3\(^ +\) and CD4\(^ +\) circulating lymphocytes was reduced in patients with XP but not in those with TTD. In this study, low natural killer (NK) activity was found in both syndromes. However, Norris et al\(^ {196}\) found reduced NK activity in patients with XP, whereas patients with TTD and CS showed normal NK activity. The authors concluded that increased susceptibility to skin cancers in XP may also be determined by reduced NK cell activity. Gaspari, Fleisher, and Kraemer\(^ {197}\) documented that lymphocytes from patients with XP have defective interferon production and may play an important role in the susceptibility to skin cancer, in addition to DNA repair defects, because NK cell function was not stimulated by interferon in patients with XP.

Ahrens et al\(^ {198}\) have documented that cells from XPD exhibit a marked UVB-induced inhibition of intercellular adhesion molecule type 1 (ICAM-1) expression after stimulation with interferon gamma. Patients with TTD showed no alteration in ICAM-1 expression, except for the TTD cells mutated at the R112H hotspot, which behave almost like XP cells.\(^ {108}\) The absence of ICAM-1 expression after irradiation inhibits skin cell interactions with lymphocytes, a process necessary for most cell-mediated immune responses and for an efficient immune surveillance in the skin, thereby further contributing to the predisposition to cancer found in patients with XP.

To explain the limited skin abnormalities of TTD, it has been speculated that scaling and thickening of ichthyotic skin in patients might shield replicative keratinocytes from UV damage.\(^ {124}\) However, we are not convinced that this mechanism has a relevant impact for cancer protection. Reduced lifespan of patients with TTD and frequent illness and lifestyle limitations in such patients may prevent them from acquiring a sufficient burden of UV radiation over time.

**TTD AS A TRANSCRIPTION DISEASE**

Clinically, it is increasingly evident that TTD with photosensitivity and CS resemble each other. Overlap in neurologic, developmental, and cutaneous abnormalities and the lack of cancer predisposition are observed. NER defects can easily explain photosensitivity and a predisposition to cancer but not growth retardation, brittle hair and nails, and neurodysmyelination as found in CS and TTD. Because for some patients the cellular responses to UV in TTD and XP are very close, it can be concluded that a pathway not involved in repair and lesion processing may be different between the two diseases. Therefore it was hypothesized that these non-XP features could be due to an impairment of basal...
transcription of some specific genes because of the dual role of the XPD and XPB helicases. In such case, nonphotosensitive patients with CS or TTD will exhibit the transcription defects, but with normal NER activity (Fig 8).

TFIIH is a high-molecular-weight protein complex with remarkable dual function in NER and initiation of RNA polymerase II transcription. TFIIH is composed of 9 subunits, in which the XBP and XPD helicases as well as the p44 protein are necessary for a normal unwinding activity at the site of transcription initiation and at the site of lesion repair (see Fig 5). All TTD lines tested appear to have smaller amounts of the TFIIH complex, which may represent the limiting factor when cells require enhanced TFIIH levels for transcription or repair, or when differentiated cells do not synthesize TFIIH de novo and amounts of this complex become suboptimal. This could explain the brittle hair/nail phenotype.

A remarkable recent finding has been the characterization of 4 unusual cases of TTD that exhibited fever-dependent, reversible deterioration of typical TTD-related features, including loss of brittle hair and worsening of ichthyosis and ataxia. One patient was a compound heterozygote in XPD with a R658C amino acid substitution in one allele and a G713R change in the other. The mutant alleles induced markedly decreased basal transcription rates in the TTD fibroblasts when subjected to elevated incubation temperatures oscillating around 41°C. Moreover, overall NER activity dropped to less than 10% of the already reduced levels seen at 37°C. This was associated with thermal instability of TFIIH. The investigators suggest that continued de novo synthesis of the complex partly compensates for the TFIIH instability until terminal differentiation is initiated (eg, in epidermis and hair follicle cells). Under these conditions with thermolabile, destabilizing TTD mutations, TFIIH is depleted more rapidly, leading to worsening of hair and skin findings with fever.

This is one of the few examples of a temperature-sensitive, heritable disorder observed in humans and characterized at the molecular level.

The dysmyelination process observed in patients with TTD may also result from insufficient transcription of abundant genes coding the structural com-
ponents of the myelin sheath. CSA and CSB genes, unlike XPB and XPD genes, are not essential for viability. Their products also interact with the RNA polymerase II, likely allowing some form of transcription regulation during RNA synthesis on the normal template and/or damaged template. This effect on transcriptional regulation may explain the resemblance of TTD with CS, but the different modes of action of these gene products on transcription may also explain the dissimilarities between the two syndromes.

This model of attenuated transcription in terminally differentiating cells in TTD has received support from studies with the fruit fly Drosophila melanogaster (Dm). The viable mutated alleles of the Dm haywire gene (the equivalent of the XPB gene product that is also part of the TFIIH complex) can cause defects in the central nervous system and growth retardation that may be akin to abnormalities in patients with CS/XP-B. Spermatogenesis in Drosophila is very sensitive to the level of the tubule protein, β2-tubulin. Mutations within the Drosophila XPB gene affect β2-tubulin expression, causing male sterility, and possibly may lower the expression of other crucial genes. It is therefore likely that expression of this gene in Drosophila, and possibly in humans, is particularly sensitive to the level of transcription at particular stages of development and sensitive to subtle mutations in protein subunits of TFIIH. This may explain the immature sexual development observed in patients with TTD and CS.

Similarly, reduced transcription of genes encoding the class of KAP 4-5 of the hair shaft may account for the reduced cysteine content in the brittle hair of patients with TTD. The typical brittle hair in TTD is, indeed, due to a reduction in the content of hair-specific cysteine-rich matrix proteins that cross-link the keratin fibers and leads to the fragile hair found in patients with TTD.

CONCLUSION

In recent years, enormous progress has been made in our understanding of the NER processes and transcription factor complexes in humans and in the molecular mechanisms underlying UV-sensitive diseases such as TTD. The constellation of growth retardation, brittle hair, and neurodysmyelination has been difficult to explain by NER defects alone. There is now strong evidence that these non-XP features of TTD are due to an impairment of the transcription function of XPD and XPB gene products, whereas the photosensitivity is a consequence of the disruption of the DNA repair function. Nonphotosensitive subtypes of TTD may be reconciled by the "repair/transcription" syndrome model in which a mutation impairs the transcription function of TFIIH but leaves the repair function intact. In normal dividing TTD cells, the abnormal TFIIH factor will be renewed sufficiently rapidly to compensate for its reduced stability. In terminally differentiated cells, however, the synthesis of de novo TFIIH may not be adequate and accumulation of inactive transcription factor and depletion may occur, giving rise to depressed basal transcription of some genes. In particular, those genes highly expressed in differentiating cells such as those involved in hair structure or in the neuromyelination process may be the targets of this disorder. Moreover, the low repair level found in cells from the photosensitive patient with TTD renders the transcription machinery even more vulnerable since RNA polymerases are blocked by DNA lesions and therefore are even less active after UV sun exposure.

The absence of skin cancers in patients with TTD may well be due to more efficient immunosurveillance than in those with XP since any premalignant or malignant cells might be targeted in TTD and, therefore, eliminated. However, taking into account results showing some predisposition to cancer in the experimental TTD mouse models, it is plausible to speculate that increased life expectancy of TTD could be accompanied by an increased risk in skin cancer. Therefore it is reasonable to recommend sun protective measures and periodic skin examinations to observe for any signs of premalignant or cancerous lesions in patients with photosensitive TTD.

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Itin, Sarasin, and Pittelkow


Directions for questions 1-28: Give single best response.

1. The term trichothiodystrophy refers to
   a. sulfur-deficient nail disease
   b. hair with methionine deficiency
   c. excessive sulfur-containing hair disorder
   d. sulfur-deficient hair with malabsorption
   e. sulfur-deficient, brittle hair syndrome

2. Trichothiodystrophy is inherited in the following pattern:
   a. autosomal dominant
   b. autosomal recessive
   c. X-linked dominant
   d. X-linked recessive
   e. paradominant

3. Which light-microscopic finding is not characteristic of trichothiodystrophy?
   a. Trichoschisis
   b. Trichorrhexis nodosa
   c. Trichorrhexis invaginata
   d. Irregular surface
   e. Irregular diameter

4. Which condition has never been associated with “dark and light banding” by polarizing microscopy?
   a. Argininosuccinic aciduria
   b. Acrodermatitis enteropathica
   c. Kwashiorkor
   d. Methionine-deficient hair
   e. Alkaptonuria

5. Which feature is not part of the Tay syndrome?
   a. Ichthyosiform erythroderma
   b. Hair shaft abnormalities
   c. Mental and somatic retardation
   d. Neutropenia
   e. Collodion baby

6. The name “Sabinas” syndrome derives from
   a. the author of the first description
   b. the name of the first patient

7. Which condition does not feature low sulfur hair content?
   a. Marinesco-Sjögren syndrome
   b. Onychotrichodysplasia
   c. Kwashiorkor
   d. Pili bifurcati
   e. Itin syndrome

8. Which disease has never been observed to harbor a DNA repair defect?
   a. BIDS syndrome
   b. Trichothiodystrophy
   c. PIBIDS syndrome
   d. Xeroderma pigmentosum
   e. Cockayne syndrome

9. Embryologically, trichothiodystrophy can best be explained as an
   a. endodermal dysplasia
   b. mesodermal dysplasia
   c. endodermal-mesodermal dysplasia
   d. ectodermal dysplasia with mesodermal and rare endodermal dysplasia
   e. pure “one-layer disease” of ectodermal origin

10. Which feature is necessary for the diagnosis of trichothiodystrophy?
    a. Somatic retardation
    b. Brittle hair
    c. Neutropenia
    d. Ichthyosis
    e. Mental retardation

11. The most common similarity between trichothiodystrophy and Cockayne syndrome is
    a. agenesis of corpus callosum
    b. central nervous system dysmyelination
    c. brittle hair and nails
thiodystrophy can be assigned to which complementation group?
   a. XPA  
   b. XPB  
   c. XPC  
   d. XPD  
   e. XPE

19. Which statement concerning the classification of trichothiodystrophy based on photosensitivity and excision repair is incorrect?
   a. Patients may show photosensitivity and no defect in excision repair of UV damage.  
   b. Patients may show no photosensitivity but a nuclear excision repair defect.  
   c. Patients may show photosensitivity and a nuclear excision repair defect in the same gene as in xeroderma pigmentosum D.  
   d. Patients may show photosensitivity and a nuclear excision repair defect in the same gene as in xeroderma pigmentosum B.  
   e. Patients may show photosensitivity and a nuclear excision repair defect in the same gene as in xeroderma pigmentosum F.

20. The gene responsible for human xeroderma pigmentosum group D has been identified as a
   a. DNA ligase  
   b. DNA helicase  
   c. catalase  
   d. hydroxylase  
   e. the gene has not yet been assigned

21. Catalase activity is
   a. increased in trichothiodystrophy compared with xeroderma pigmentosum  
   b. increased in xeroderma pigmentosum compared with trichothiodystrophy  
   c. decreased in trichothiodystrophy  
   d. equivalent in xeroderma pigmentosum and trichothiodystrophy  
   e. similar in xeroderma pigmentosum compared with normal controls

22. The human ERCC2/XPD gene maps to the long arm of
   a. chromosome 2  
   b. chromosome 6  
   c. chromosome 7  
   d. chromosome 12  
   e. chromosome 19

23. The XPD/ERCC2 gene is involved in
   a. recognition of DNA lesions  
   b. unwinding DNA in the vicinity of a lesion  
   c. ligation of DNA strands  
   d. DNA base substitution  
   e. gap filling by DNA synthesis

24. The most common mutation identified in photosensitive patients with trichothiodystrophy is
   a. Arg112His substitution  
   b. Leu461Val substitution  
   c. Arg683Trp substitution
d. Arg722Try substitution
e. Arg658His substitution

25. Predisposition to cancer is favored in xeroderma pigmentosum but not in trichothiodystrophy because
   a. catalase activity is not decreased in trichothiodystrophy.
   b. no alteration of intercellular adhesion molecule 1 expression is present in trichothiodystrophy but is present in xeroderma pigmentosum.
   c. p53 tumor suppressor gene product is enhanced after UV radiation in xeroderma pigmentosum but not in trichothiodystrophy.
   d. CD3 and CD4 lymphocytes are reduced in trichothiodystrophy.
   e. None of the above-mentioned mechanisms are correct.

26. What sign or symptom is not observed in mice with trichothiodystrophy?
   a. Brittle hair
   b. Developmental abnormalities
   c. Neutropenia
   d. UV sensitivity
   e. Skin abnormalities

27. The mice with trichothiodystrophy were obtained by
   a. spontaneous mutation
   b. radiation mutagenesis
   c. chemical mutagenesis
   d. transgenic procedure
   e. mating of an ichthyotic mouse with the hairless mouse

28. What is the most important difference between the mouse model of trichothiodystrophy and the human disease?
   a. Normal hair in the mouse
   b. Increased cancer risk from chemical carcinogens in the mouse
   c. No other developmental abnormalities
   d. Normal life span in mice
   e. Normal size compared with control mice

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Answers to CME examination

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1. e
2. b
3. c
4. e
5. d
6. c
7. d
8. a
9. d
10. b
11. b
12. b
13. e
14. a
15. b
16. d
17. c
18. d
19. e
20. b
21. a
22. e
23. b
24. a
25. a
26. c
27. d
28. b

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