dase subunit present but oxidaitive burst activity is not at a level sufficient to fight off infection.

In 1986, the gp91phox subunit was first cloned [27], encoded from the CYBB gene, although little homology with other known protein sequences shed inadequate light on its function at the time. This was a huge advance in the molecular understanding of CGD and led to the development of animal models and the ability to determine what controls gp91phox function and activity; as well as being fundamental to current gene therapy development. Subsequently, by screening a promyelocytic leukemia cDNA library, Volpp *et al.* (1989) cloned and sequenced a cDNA encoding the 47-kD component of the NADPH oxidase system [28], Leto *et al.* (1990) [29] cloned a p67phox cDNA while Dinauer *et al.* [30] reported the structure of the gene for the 22-kD light chain of cytochrome-b_{558} and its chromosomal location, which served as a foundation for the analysis of genetic abnormalities at this locus in CGD. The cloning of p40phox by Wientjes *et al.* [31] provided important insights into its interactions with p67phox and p47phox in the cytosol. It was first suspected in the late 1980s that a GTPase might play a role in NADPH oxidase activation when it was demonstrated that guanine nucleotides were able to stimulate oxidase activity. The GTPase was subsequently identified and cloned as Rac1 or Rac2 and it is now clear that its presence is absolutely required for full oxidase function [32].

Mutations in the X-linked and autosomal recessive forms of CGD

Research into CGD has improved our knowledge of the normal NADPH oxidase system. In some cases, the identification of specific mutations has provided insights at a molecular level. Most patients with CGD have mutations in the CYBB gene that encodes gp91phox, located at Xp21.1. Since the discovery of the CYBB gene no single mutation appeared to be responsible and a flood of research on mutations ensued, reporting large and small deletions, frameshifts and other mutations. The genetics of many hundreds of families with CGD were investigated and recounted and a mutation registry database for X-linked and autosomal recessive CGD was set up by Dirk Roos and MaunoVihinen from the University of Tampere in Finland [33] which presently lists 304 mutation entries from 267 unrelated families showing about 204 unique molecular events. Most of the X-linked mutations (174) are in the N-terminal domain. The number of mutations in other domains report: 50 in the FAD domain, 49 in the NADPH domain, 7 in the loop region, 3 in the upstream, the non-coding region before initiation codon, and 20 undefined gross deletions. The most common mutations are nonsense mutations (81), followed by missense mutations (78), frameshift deletions (42), intron mutations (34), frameshift insertions (30), and inframe deletions (6) [33,34].

Autosomal recessive CGD arises from mutations occurring in either the p67phox, p47phox or p21phox phagocyte oxidase proteins. Mutations in the NCF2 gene which encodes the p67phox represents about 5% of CGD cases whereas deficiencies of NCF1 which encodes p47phox represents about a quarter of all cases of CGD [35]. Unlike other CGD subtypes, in which there is great heterogeneity among mutations, 97% of affected alleles in patients with p47phox deficiency carry a characteristic 2-base pair deltaGT deletion in the NCF1 gene [36]. p22phox mutations are less common and 2 novel nonsense and missense mutations in the CYBA gene have been described [37]. Mutations in the corresponding genes responsible for the different genetic subgroups of CGD are shown in Table 1. Genetic defects in Rap1a, Rac, or p40phox havenot been reported in CGD [38], however, a dominant-negative mutation in the hematopoietic-specific Rac2 GTPase was identified in an infant with severe neutrophil dysfunction and a predisposition to bacterial infections similar to CGD and leukocyte adhesion deficiency [39].

Activation of the NADPH oxidase complex

The dormant oxidase consists of both cytosolic and membrane-bound components, but the active $O2^{-\cdot}$ generating complex is confined to the plasma membrane [1,2]. This implies that the cytosolic components must either act in a signaling capacity by modifying the membrane components, or that they must become directly associated with the membrane via a translocation process.

The 47 kDa phosphoprotein was found in both the cytosol and membranes after stimulation of neutrophils with phorbol myristate acetate and the phosphoprotein

Table 1: Classification of CGD subunits.

CGD gene	Genetic Transmission	Frequency (%)
gp91phox (CYBB)	X-linked	65
p47phox (NCF1)	Autosomal recessive	25
p67phox (NCF2)	Autosomal recessive	5
p22phox (CYBA)	Autosomal recessive	5

Mutations can occur in one of four NADPH oxidase subunits which give rise to X-linked or autosomal recessive CGD. The approximate frequencies of case occurrence are shown, with figures extrapolated from Clark *et al.* [35] and Dineaur *et al.*[38].