M. Sekiguchi: Molecular Mechanisms for Controlling Mutations in Mammalian Cells

Spont. Mutagenesis in Ecol,

Pre-replication — Substrate mut T; Template mut M, mut Y

↓

Replication — Polymerase dnaE; Editing dnaQ

↓

Post replication — Mismatch mut S, mut L, mut H, uvrD

Mut T

Treffers et al. — isolated mutant

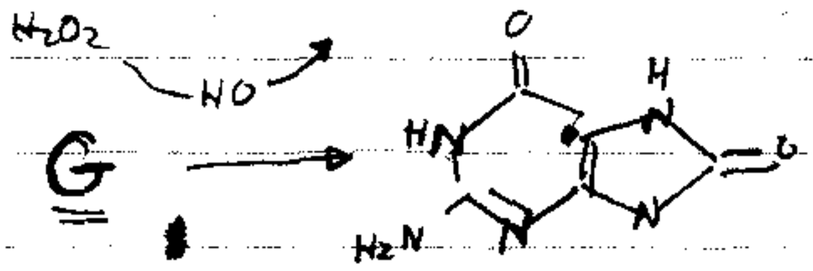
Yanofsky ... mut T leads to AT → GG transversion

Akiyama — mut T gene

Maki & Sekiguchi — mut T = 8-oxodGTPase

(leads to %GC increase)

mw = 15 kDa



8-oxo-dG

mut T
dC:dA-O-G mut M = 8-oxo-dG glycosylase
dA:dA-O-G mut Y = adenine glycosylase

mut T mut M = GG→AT Δs

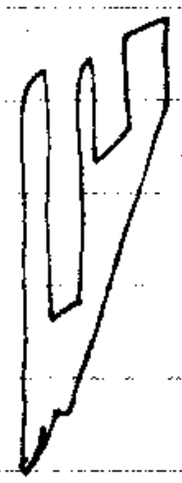
-C-
-G-
↓
-C-
-G-
x

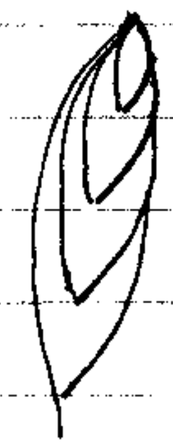
human 8-oxo-dGTPase

- purified activity
- peptide sequence
- cloned cDNA
- express in E coli = activity

E coli	mutT like
Proteus	"
Streptococcus	"
rat	"
mouse	"
human	" = chromosome 7

① search for MTH1 mutants
② creation of MTH1 KO'd mice





O-6-MeG-Metransferase

: disrupt mouse MGMT
: -/- slower growing than +/+
: -/- MNU sensitive