Table 9.6 Modifications related to ribose and backbone.

Problem	Affected locations	Mass difference (Ave.)	Source
+PS (phosphorothioate)	Backbone	+96.05	Process
+PO (phosphate)	Backbone	+79.98	Process
PS -> PO conversion	Backbone	-16.07	Process (capping, deprotection, oxidation)
Phosphorodithioate instead of phosphorothioate	Backbone	+16.07	Process (thiolation)
Cyclic phosphate (3'-truncation)	Backbone, ribose	+61.96	Process (deprotection)
+Chloral or +trichloroacetaldehyde	Backbone	+147.39	Raw material (DCA)
+2'-*O*-tert-butyl dimethylsilyl (TBDMS)	Ribose at rA, rG, rC, U	+114.26	Process (deprotection)
+DMT-C phosphonate	Internucleotide bond	+286.37	Detritylation
+DMT-C phosphonate	5'-Terminal phosphate	+366.10	Detritylation
+DMT	5'-*O*-position	+302.37	Process (detritylation)
-HF	2-F' pyrimidines	-19.99	Process (deprotection); storage
-HF, $+H_2O$	2-F' pyrimidines	-2.00	Process (deprotection); storage
2'/3'-Isomer	Ribose; any RNA or 2'-mod RNA nucleoside	±0.00	Raw material
Internucleotide linkage (3'-2'-migration)	Ribose: any RNA nucleoside		Process (deprotection)
Inverted base (5'-3'-inversion)	Ribose: any RNA or 2'-mod. RNA nucleoside		Raw material
2'-*O*-methyl	Any MOE nucleoside	-44.05	Raw material
2'-*O*-propyl	Any MOE nucleoside	-16.00	Raw material
2'-*O*-butyl	Any MOE nucleoside	-1.82	Raw material
2'-*O*-(2-ethoxy-ethyl), EOE	Any MOE nucleoside	+14.03	Raw material

Table 9.7 Modifications related to individual bases. Average mass differences relative to the full-length product for common impurities.

Problem	Affected locations	Mass difference (Ave.)	Source
Branch-mer (amino group of base)	A, C, or G base	*n*+ (*n*–*x*)	Loss of protecting group
Branch-mer (O^6 of G-base)	G or I base	*n*+ (*n*–*x*)	Unprotected O^6-position
Depurination (abasic site)	A base	-117.12	Process (detritylation)
Depurination (gas phase)	A base	-135.13	Instrument-induced
Depurination (abasic site)	G base	-133.11	Process (detritylation)
Ribonolactone	G base	-135.13	Process (detritylation)
Depurination (gas phase)	G base	-151.13	Instrument-induced
Depurination, ethoxyacetal-formation	G base	-105.06	Process (deprotection)
Depurination, ethoxyacetal-formation	A base	-89.06	Process (deprotection)
Deamination	5-Me-C, C, A, G base	+0.98	Process (deprotection)
2,6 Diaminopurine	G base	-0.98	Process (capping)
Transamination	C base; amino linker	+14.03	Process (deprotection)
Protection group exchange; acetyl remains on functional groups	G base; amino linker	+42.04	Process (capping; deprotection)
+b-*N*-methylamino acetamide (MAM)	Unknown	+85.10	Raw material (capping)
N_2-acetyl-2,6-diaminopurine (ADP)	G base	+41.05	Process (capping)
N_2-isobutyryl-2,6-diaminopurine (IDP)	G base	+69.11	Process (capping)
3-(3-Acetyl-4-methylpyridine-2-one-6-yl)-2-aminoimidazole (AMPA)	A base	+98.04	Process (capping)
+Cyanoethyl (CNET)	Heterocyclic bases, predominantly T-base	+53.06	Process (deprotection)
Residual protection group; +isobutyryl	^{ibu}G base	+70.08	Process (deprotection)
Residual protection group; +benzoyl	^{bz}A base	+104.11	Process (deprotection)
3,N^6-Etheno-cytidine derivative	C base	+80.08	Process, thermal stress
Depyrimidation (abasic side)	U base	-52.03	Process (deprotection)
Depyrimidation (abasic side)	U base	-94.07	Process (deprotection)
8-Oxo-formation	A, G-base	+16.00	Process (oxidative stress)

Table 9.7 (continued)

Problem	Affected locations	Mass difference (Ave.)	Source
5-Hydroxymethyl cytosine	5-MeC-base	+16.00	Process (oxidative stress)
±dA (PO)	Molecule integrity	±313.21	Process (synthesis)
±dG (PO)	Molecule integrity	±329.21	Process (synthesis)
±dC (PO)	Molecule integrity	±289.18	Process (synthesis)
±T (PO)	Molecule integrity	±304.19	Process (synthesis)
±rA (PO)	Molecule integrity	±329.21	Process (synthesis)
±rG (PO)	Molecule integrity	±345.21	Process (synthesis)
±rC (PO)	Molecule integrity	±305.18	Process (synthesis)
±U (PO)	Molecule integrity	±306.17	Process (synthesis)
±2'O-Me-A (PO)	Molecule integrity	±343.23	Process (synthesis)
±2'O-Me-G (PO)	Molecule integrity	±359.23	Process (synthesis)
±2'O-Me-C (PO)	Molecule integrity	±319.21	Process (synthesis)
±2'O-Me-U (PO)	Molecule integrity	±320.19	Process (synthesis)
±2'F-dA (PO)	Molecule integrity	±331.20	Process (synthesis)
±2'F-dG (PO)	Molecule integrity	±347.20	Process (synthesis)
±2'F-dC (PO)	Molecule integrity	±307.17	Process (synthesis)
±2'F-dU (PO)	Molecule integrity	±308.16	Process (synthesis)
±LNA-A (PO)	Molecule integrity	±341.22	Process (synthesis)
±LNA-G (PO)	Molecule integrity	±357.22	Process (synthesis)
±LNA-C (PO)	Molecule integrity	±317.19	Process (synthesis)
±LNA-U (PO)	Molecule integrity	±318.18	Process (synthesis)
±2'-MOE-A (PO)	Molecule integrity	±387.29	Process (synthesis)
±2'-MOE-G (PO)	Molecule integrity	±403.29	Process (synthesis)
±2'-MOE-5MeC (PO)	Molecule integrity	±377.29	Process (synthesis)
±2'-MOE-5MeU (PO)	Molecule integrity	±378.27	Process (synthesis)

Table 9.8 Unylinker-related impurities.

Problem	Affected locations	Mass difference (Ave.)	Source
Unylinker, dimer impurity PO (PS)	3’-end; Unylinker	+442.33 (+458.33)	Raw material
Unylinker, dimer impurity PO (PS)	3’-end; Unylinker	+400.35 (+476.35)	Raw material
Unylinker, dimer impurity PO (PS)	3’-end; Unylinker	+478.36 (+494.36)	Raw material
Unyliner, partial deprotection, PO (PS)	3’-end; Unylinker	+261.13 (+277.13)	Process (deprotection)
Unyliner, partial deprotection, PO (PS)	3’-end; Unylinker	+275.15 (+281.15)	Process (deprotection)
Unyliner, partial deprotection, PO (PS)	3’-end; Unylinker	+278.16 (+294.16)	Process (deprotection)
Unyliner, partial deprotection, PO (PS)	3’-end; Unylinker	+368.28 (+384.28)	Process (deprotection)
Unyliner, partial deprotection, PO (PS)	3’-end; Unylinker	+354.06 (+370.06)	Process (deprotection)