MW and TM calculation

Oligo Specification Report accompanying all custom oligo synthesis including ALL modifications uses the molecular weights given below.

The molecular weights are not added for modifications and backbone modifications.

ID	Base	BaseAbbreviation	MW	EC	Notes
1	DeoxyAdenosine	A	313.21	15.4	
2	DeoxyCytosine	C	289.19	7.4	
3	DeoxyGuanosine	G	329.21	11.5	
4	Thymidine	T	304.2	8.7	
5	Inosine	I	314.2	7.2	
6	A+G+T+C	N	308.95	10.7	
7	A+G	R	321.21	13.45	
8	C+T	Y	296.69	8.05	
9	A+C	M	301.2	11.4	
10	G+T	K	316.7	10.1	
11	G+C	S	309.2	9.45	
12	A+T	W	308.71	12.05	
13	A+T+C	H	302.2	10.5	
14	G+T+C	B	307.53	9.2	
15	G+A+T	D	315.54	11.86	
16	G+A+C	V	310.53	11.43	
18	phosphate	p	79.98	0	
19	Other	X	0	0	add mw of the
20	deoxy uridine	U	290.17	9.9	

Formulas

Size= Total number of bases.

%GC= (G+C)/Size

mw = (A x 313.2) + (C x 289.19) + (G x 329.21) + (T x 304.2) + (I x 314.2) + (N x 308.95) + (R x 321.21) + (Y x 296.69) + (M x 301.2) + (K x 316.7) + (S x 309.2) + (W x 308.71) + (H x 302.2) + (B x 307.53) + (D x 315.54) + (V x 310.53) + (P x 79.98) + (U x 290.17) –62

Tm For Oligos shorter than 25 bp = 2(A+T) + 4 (C+G)

For longer oligos: Reference Bolton, Et and McCarthy, B.J. (1962) PNAS 48: 139-1397

Tm=81.5 – 16.6 + (0.41 x %GC)) – 600 / size

At Gene Link the website online ordering software and the oligo report enclosed with the shipment uses the following formula for calculation of melting temperature (Tm) of the oligonucleotide

$$T_m = 81.5 + 16.6 \times \text{Log}_{10}[Na^+] + 0.41 \times P_{GC} - 600/N$$

Where: P_{GC} is the total percentage of G and C bases
N is the sequence length
$[Na^+]$ is 100 mM

Example: 5'- TATATGCCCGATCCGCTAGT-3' 20 mer; GC = 50%; AT = 50%

$T_m = 81.5 + 16.6 \times \text{Log}_{10}[Na^+] + 0.41 \times P_{GC} - 600/N$

$T_m = 81.5 + 16.6 \times \text{Log}_{10}[0.100] + 0.41 \times 50 - 600/20$

$T_m = 81.5 - 16.6 + \underline{0.41 \times 50} - \underline{600/20}$

$T_m = 81.5 - 16.6 + 20.5 - 30$

$T_m = 64.9 + 20.5 - 30$

$T_m = 85.40 - 30$

$T_m = 55.4^oC$

Basic

$T_m = (2 \times N_{AT}) + (4 \times N_{GC})$

Where:
N_{AT} is the total number of A or T bases
N_{GC} is the total number of C or G bases

Tm for same oligo using 2(A+T) + 4 (C+G)

= 2(5+5) + 4(5+5)
= 2(10) + 4(10)
= 20+ 40
= 60 ^{o} C

DNA Extinction Coefficients

Nucleotide	(A260 / mol)
A	15.4
T	8.7
G	11.5
C	7.4
I	7.2**
N	10.7*

Neighbors	(A260 / mol)
AA	13.7
AT	11.4
AG	12.5
AC	10.6
AI	9.3**
AN	12.2*
TA	11.7
TT	8.4
TG	9.5
TC	8.1
TI	8.1**
TN	9.4*
GA	12.6
GT	10.0
GG	10.8
GC	8.8
GI	8.8**
GN	10.5*
CA	10.6
CT	7.6
CG	9.0
CC	7.3
CI	7.2**
CN	8.6*
IA	9.3**
IT	8.4**
IG	8.8**
IC	7.1**
II	6.8**
IN	8.4*
NA	12.1*
NT	9.4*
NG	9.4*
NC	8.7*
NI	8.7*
NN	9.9*

Handbook of Biochemistry and Molecular Biology (1975) Fasman G.D., ed., 3rd edition, Nucleic Acids - Vol. 1, pp 589, CRC Press, Cleveland, OH.