

Tesi Doctoral

**Formació del motiu quàdruplex *bi-loop* amb oligonucleòtids lineals
i aplicació a la ciclació assistida per motlle**

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ANNEX



ABREVIATURES, ACRÒNIMS I SÍMBOLS

A, Ade	adenina
Abs	absorbància
ACN	acetonitril
anh.	anhidre/a
AP	fosfatasa alcalina
Ar	aromàtic
ATP	adenosina trifosfat
bp	parells de bases (<i>base pairs</i>)
C, Cyt	citosina
CA	citrat d'amoni
CCF	cromatografia en capa fina
CNE	2-cianoetil
CPG	suport de boles de vidre de porus controlat (<i>Controlled Pore Glass</i>)
CPR	reactiu fosforilant (<i>Chemical Phosphorylation Reagent</i>)
dA	2'-desoxiadenosina
dC	2'-desoxicitidina
DC	dicroisme circular
DCC	<i>N,N'</i> -diciclohexilcarbodiimida
DCM	diclorometà
dG	2'-desoxiguanosina
DIEA	<i>N,N'</i> -diisopropiletilamina
dT	timidina
COSY	<i>CORrelated SpectroscopY</i>
diOBt	fosfat de bis-(1-benzotriazolil) i 2-clorofenil
DMF	<i>N,N</i> -dimetilformamida
DMT	4,4'-dimetoxitritil
dN	2'-desoxiribonucleòtid
DNA	àcid desoxiribonucleic
dsDNA	àcid desoxiribonucleic de doble cadena (<i>double stranded DNA</i>)
DSS	2,2-dimetil-2-silapentà-5-sulfonat de sodi perdeuterat
DTS	síntesi orgànica assistida per DNA (<i>DNA-templated organic synthesis</i>)
EDC	<i>N</i> -(3-dimetilaminopropil)- <i>N'</i> -etilcarbodiimida
EDTA	àcid etilendiaminotetraacètic
eq.	equivalents
ESI	espectrometria de masses per ionització electrosprai
f	funcionalització (grau de substitució de la resina)
Fmoc	9-fluorenilmetoxicarbonil
G, Gua	guanina
HOBt	1-hidroxibenzotriazole
HPA	àcid 3-hidroxipicolínic
HPLC	cromatografia líquida d'alta eficàcia (<i>High Performance Liquid Chromatography</i>)
Icaa	cadena llarga d'alquilamina (<i>long chain alkylamino</i>)
M	massa monoisotòpica
MALDI-TOF	espectrometria de masses de desorció iònica provocada per làser i assistida per matriu amb analitzador de temps de vol (<i>Matrix Assited Laser Desorption Ionization-Time of Flight</i>)
mdeg	miligras (<i>milidegrees</i>)
MES	àcid morfolinoetansulfònic
MPLC	cromatografia líquida de mitja resolució (<i>Medium Performance Liquid Chromatography</i>)
mRNA	àcid ribonucleic missatger
MS	espectrometria de masses (<i>Mass Spectrometry</i>)
MSNT	1-mesitilensulfonil-3-nitro-1,2,4-triazole
m/z	relació massa/càrrega
Na₂PIPES	1,4-piperazina-bis(etansulfonat) de sodi

NMI	<i>N</i> -metilimidazole
NOE	<i>Nuclear Overhauser Effect</i>
NOESY	<i>Nuclear Overhauser Effect Spectroscopy</i>
nt	nucleòtid
OD₂₆₀	densitat òptica a 260 nm
P	fosfat
PAGE	electroforesi en gel de poliacrilamida (<i>PoliAcrylamide Gel Electrophoresis</i>)
PCR	reacció en cadena de la polimerasa (<i>Polymerase Chain Reaction</i>)
PEG	polietilenglicol
P_f	punt de fusió
Pip	piperidina
PM	pes molecular
PNA	àcid nucleic peptídic (<i>Peptide Nucleic Acid</i>)
PS	poliestirè-co-1%-divinilbenzè
Pir	piridina
ppm	parts per milió
Pur, R	purina
Pyr, Y	pirimidina
R	suport polimèric o resina
RCA	mecanisme del cercle rodant (<i>Rolling Circle Mechanism</i>)
RMD	Dinàmica Molecular Restringida (<i>Restrained Molecular Dynamics</i>)
R_f	factor de retenció
RMN	espectroscòpia de Ressonància Magnètica Nuclear
RMSD	desviació quadràtica mitjana (<i>Root Mean Square Deviation</i>)
RNA	àcid ribonucleic
ROESY	<i>Rotating frame nuclear Overhauser Effect Spectroscopy</i>
RX	crystal·lografia de Raigs X
SIR	<i>selected (or single) ion recording</i>
SpPD	fosfodiesterasa de melsa de bou (<i>Bovine Spleen Phosphodiesterase</i>)
ssDNA	àcid desoxiribonucleic de cadena senzilla
SVPD	fosfodiesterasa de verí de serp (<i>Snake Venom Phosphodiesterase</i>)
stains-all-dye	4,5,4',5'-dibenzo-3,3'-diètil-9-metilcarbocianat
T, Thy	timina
T_{am}	temperatura ambient
TBAF	fluorur de tetrabutilamoni
TBE	tampó Tris borat EDTA
TCA	àcid tricloroacètic
TDS	espectre de diferència tèrmica (<i>Thermal Difference Spectrum</i>)
TEA	triètilamina
TEMED	<i>N,N,N',N'</i> -tetrametilètilendimaina
Tet	tetrazole
THAP	2,4,6-trihidroxiacetofenona
THF	tetrahidrofurà
T_m	temperatura de fusió d'una estructura de DNA (°C)
TMG	<i>N,N,N',N'</i> -tetrametilguanidina
TMP	fosfit de trimetil
TMS	trimetilsilà
t_R	temps de retenció (en HPLC)
Tris	tris(hidroximetil)aminometà
TSP	sal sòdica de l'àcid 3-(trimetilsilil)-propioníc perdeuterat
TOCSY	<i>TOTAL Correlation Spectroscopy</i>
U	unitat
UV-Vis	espectroscòpia d'ultravioleta-visible
WC	Watson-Crick (parells canònics)
wg	<i>watergate</i> (en els espectres d'RMN, forma d'eliminar la senyal de l'aigua)

CICLES DE SÍNTESI D'OLIGONUCLEÒTIDS

Cicle ce102a: cicle estàndard sobre CPG a escala 1 µmol en ABI380B.

STEP NUMBER	FUNCTION # NAME	STEP TIME	STEP ACTIVE FOR BASES							SAFE STEP	
			A	G	C	T	5	6	7		
1	83 #13 To Waste	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
2	15 #13 To Column	60	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
3	2 Reverse Flush	20	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
4	1 Block Flush	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
5	28 Phos Prep	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
6	+45 Group 1 On	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
7	90 TET to column	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
8	19 B+TET To Col 1	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
9	90 TET to column	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
10	19 B+TET To Col 1	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
11	90 TET to column	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
12	19 B+TET To Col 1	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
13	90 TET to column	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
14	-46 Group 1 Off	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
15	4 Wait	30	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
16	16 Cap Prep	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
17	10 #18 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
18	2 Reverse Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
19	1 Block Flush	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
20	+45 Group 1 On	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
21	22 Cap To Col 1	20	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
22	-46 Group 1 Off	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
23	4 Wait	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
24	10 #18 To Waste	7	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
25	2 Reverse Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
26	1 Block Flush	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
27	81 #15 To Waste	7	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
28	13 #15 To Column	23	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
29	4 Wait	30	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
30	10 #18 To Waste	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
31	2 Reverse Flush	20	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
32	1 Block Flush	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
33	9 #18 To Column	20	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
34	2 Reverse Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
35	9 #18 To Column	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
36	2 Reverse Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
37	9 #18 To Column	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
38	2 Reverse Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
39	9 #18 To Column	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
40	2 Reverse Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
41	1 Block Flush	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
42	33 Cycle Entry	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
43	10 #18 To Waste	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
44	9 #18 To Column	20	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
45	2 Reverse Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
46	1 Block Flush	6	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
47	6 Waste-Port	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
48	5 Advance FC	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
49	82 #14 To Waste	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
50	14 #14 To Column	50	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
51	1 Block Flush	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
52	10 #18 To Waste	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
53	9 #18 To Column	120	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
54	2 Reverse Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
55	1 Block Flush	4	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
56	7 Waste-Bottle	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

Cicle MSPEGPSM: cicle de síntesi d'oligonucleòtids a escala mitjana sobre TentaGel en ABI380B.

STEP NUMBER	FUNCTION # NAME	STEP TIME	STEP ACTIVE FOR BASES							SAFE STEP
			A	G	C	T	5	6	7	
1	83 #13 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
2	15 #13 To Column	45	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
3	2 Reverse Flush	45	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
4	28 Phos Prep	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
5	+45 Group 1 On	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
6	90 TET to column	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
7	19 B+TET To Col 1	25	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
8	-46 Group 1 Off	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
9	4 Wait	900	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
10	10 #18 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
11	9 #18 To Column	30	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
12	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
13	16 Cap Prep	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
14	+45 Group 1 On	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
15	22 Cap To Col 1	30	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
16	-46 Group 1 Off	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
17	4 Wait	120	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
18	10 #18 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
19	9 #18 To Column	45	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
20	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
21	81 #15 To Waste	7	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
22	13 #15 To Column	30	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
23	4 Wait	60	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
24	18 #16 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
25	12 #16 To Column	60	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
26	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
27	30 #17 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
28	11 #17 To Column	60	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
29	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
30	10 #18 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
31	9 #18 To Column	30	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
32	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
33	9 #18 To Column	30	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
34	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
35	33 Cycle Entry	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
36	10 #18 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
37	9 #18 To Column	20	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
38	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
39	6 Waste-Port	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
40	5 Advance FC	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
41	82 #14 To Waste	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
42	14 #14 To Column	120	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
43	1 Block Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
44	10 #18 To Waste	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
45	9 #18 To Column	200	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
46	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
47	1 Block Flush	5	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
48	7 Waste-Bottle	1	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
49	30 #17 To Waste	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
50	11 #17 To Column	30	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
51	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
52	10 #18 To Waste	3	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
53	9 #18 To Column	20	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
54	2 Reverse Flush	10	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

Cicle DNA 1 μmol : cicle estàndard sobre CPG a escala 1 μmol en Expedite.

```

/* ----- */
/*      Function                Mode  Amount  Time(sec)  Description                */
/*      /Arg1   /Arg2                                     */
/* ----- */
$Deblocking
144 /*Index Fract. Coll.  */ NA      1      0  "Event out ON"
0 /*Default                */ WAIT    0      1.5 "Wait"
141 /*Trityl Mon. On/Off  */ NA      1      1  "START data collection"
16 /*Dblk                  */ PULSE  10     0  "Dblk to column"
16 /*Dblk                  */ PULSE  50     49  "Deblock"
38 /*Diverted Wsh A       */ PULSE  40     0  "Flush system with Wsh A"
141 /*Trityl Mon. On/Off  */ NA      0      1  "STOP data collection"
38 /*Diverted Wsh A       */ PULSE  40     0  "Flush system with Wsh A"
144 /*Index Fract. Coll.  */ NA      2      0  "Event out OFF"
$Coupling
1 /*Wsh                    */ PULSE  5      0  "Flush system with Wsh"
2 /*Act                    */ PULSE  5      0  "Flush system with Act"
18 /*A + Act               */ PULSE  5      0  "Monomer + Act to column"
18 /*A + Act               */ PULSE  2     16  "Couple monomer"
2 /*Act                    */ PULSE  3     24  "Couple monomer"
1 /*Wsh                    */ PULSE  7     56  "Couple monomer"
1 /*Wsh                    */ PULSE  8      0  "Flush system with Wsh"
$Capping
12 /*Wsh A                 */ PULSE  20     0  "Flush system with Wsh A"
13 /*Caps                  */ PULSE  8      0  "Caps to column"
12 /*Wsh A                 */ PULSE  6     15  "Cap"
12 /*Wsh A                 */ PULSE  14     0  "Flush system with Wsh A"
$Oxidizing
15 /*Ox                    */ PULSE  15     0  "Ox to column"
12 /*Wsh A                 */ PULSE  15     0  "Flush system with Wsh A"
$Capping
13 /*Caps                  */ PULSE  7      0  "Caps to column"
12 /*Wsh A                 */ PULSE  30     0  "End of cycle wash"

```

Cicle DNA 5 μmol TentaGel: cicle de síntesi d'oligonucleòtids a escala mitjana sobre TentaGel en Expedite.

```

/* ----- */
/*      Function                Mode  Amount  Time(sec)  Description                */
/*      /Arg1   /Arg2                                     */
/* ----- */
$Deblocking
144 /*Index Fract. Coll.  */ NA      1      0  "Event out ON"
0 /*Default                */ WAIT    0      1.5 "Wait"
141 /*Trityl Mon. On/Off  */ NA      1      1  "START data collection"
16 /*Dblk                  */ PULSE  25     0  "Dblk to column"
16 /*Dblk                  */ PULSE  150    120  "Deblock"
38 /*Diverted Wsh A       */ PULSE  120    0  "Flush system with Wsh A"
141 /*Trityl Mon. On/Off  */ NA      0      1  "STOP data collection"
38 /*Diverted Wsh A       */ PULSE  120    0  "Flush system with Wsh A"
144 /*Index Fract. Coll.  */ NA      2      0  "Event out OFF"
$Coupling
1 /*Wsh                    */ PULSE  60     0  "Flush system with Wsh"
7 /*g                      */ PULSE  60     0  "g to column"
1 /*Wsh                    */ PULSE  160    0  "Flush system with Wsh"
2 /*Act                    */ PULSE  25     0  "Flush system with Act"
18 /*A + Act               */ PULSE  15     0  "Monomer + Act to column"
18 /*A + Act               */ PULSE  25    600  "Couple monomer"
2 /*Act                    */ PULSE  10    100  "Couple monomer"
1 /*Wsh                    */ PULSE  20     60  "Couple monomer"
1 /*Wsh                    */ PULSE  60     0  "Flush system with Wsh"
$Capping
12 /*Wsh A                 */ PULSE  80     0  "Flush system with Wsh A"
13 /*Caps                  */ PULSE  25     0  "Caps to column"
13 /*Caps                  */ PULSE  40    120  "Caps to column"
12 /*Wsh A                 */ PULSE  10     20  "Cap"
12 /*Wsh A                 */ PULSE  80     0  "Flush system with Wsh A"
$Oxidizing
15 /*Ox                    */ PULSE  25     0  "Ox to column"
15 /*Ox                    */ PULSE  40     60  "Ox to column"
12 /*Wsh A                 */ PULSE  240    0  "Flush system with Wsh A"

```

Cicle MSNT 5 μ mol (1 column): cicle especial per a dur a terme la ciclació amb MSNT en pir en el sintetitzador Expedite (una única columna).

```

/* -----
/*      Function                Mode Amount Time(sec)      Description
/*                               /Arg1 /Arg2
/* -----
$Deblocking
38 /*Diverted Wsh A          */ PULSE    25     0  "Diverted Wsh A"
$Coupling
1 /*Wsh                      */ PULSE    60     0  "ACN anh"
5 /*7                        */ PULSE    60     0  "pyr anh"
6 /*8                        */ PULSE    30     0  "MSNT "
6 /*8                        */ PULSE    20    900  "MSNT "
0 /*Default                  */ WAIT     0    900  "wait"
6 /*8                        */ PULSE    20    900  "MSNT "
0 /*Default                  */ WAIT     0    900  "wait"
6 /*8                        */ PULSE    20    900  "MSNT "
0 /*Default                  */ WAIT     0    900  "wait"
6 /*8                        */ PULSE    20    900  "MSNT"
0 /*Default                  */ WAIT     0    900  "wait"
5 /*7                        */ PULSE    40     0  "pyr anh"

```

Cicle MSNT 5 μ mol (2 column): cicle especial per a dur a terme la ciclació amb MSNT en pir en el sintetitzador Expedite (2 columnes en paral·lel).

```

/* -----
/*      Function                Mode Amount Time(sec)      Description
/*                               /Arg1 /Arg2
/* -----
$Deblocking
38 /*Diverted Wsh A          */ PULSE    10     0  "Diverted Wsh A"
$Coupling
1 /*Wsh                      */ PULSE    30     0  "ACN anh"
5 /*7                        */ PULSE    30     0  "pyr anh"
6 /*8                        */ PULSE    14     0  "MSNT "
$Capping
0 /*Default                  */ WAIT     0    35  "Default"
$Coupling
6 /*8                        */ PULSE    5    900  "MSNT "
$Capping
0 /*Default                  */ WAIT     0     5  "wait"
$Coupling
6 /*8                        */ PULSE    5    900  "MSNT "
$Capping
0 /*Default                  */ WAIT     0     5  "wait"
$Coupling
6 /*8                        */ PULSE    5    900  "MSNT "
$Capping
0 /*Default                  */ WAIT     0     5  "Default"
$Coupling
5 /*7                        */ PULSE    40     0  "pyr anh"
6 /*8                        */ PULSE    14     0  "MSNT "
$Capping
0 /*Default                  */ WAIT     0    35  "Default"
$Coupling
6 /*8                        */ PULSE    5    900  "MSNT "
$Capping
0 /*Default                  */ WAIT     0     5  "wait"
$Coupling
6 /*8                        */ PULSE    5    900  "MSNT "
$Capping
0 /*Default                  */ WAIT     0     5  "wait"
$Coupling
6 /*8                        */ PULSE    5    900  "MSNT "
$Capping
0 /*Default                  */ WAIT     0     5  "Default"
$Coupling
5 /*7                        */ PULSE    40     0  "pyr anh"

```

Cicle 1um-PO: cicle estàndard sobre CPG a escala 1 µmol en ABI3400.

```

#####
## Tet times for pushing amidite to each column

TTIME 1      3.9
TTIME 2      4.1
TTIME 3      4.3
TTIME 4      4.5

#####
## Couple time for each base, and a default time for those not specified

CTIME A      30
CTIME G      30
CTIME C      30
CTIME T      30
CTIME 5      300
CTIME 6      300
CTIME 7      300
CTIME 8      300
CTIME Default 20

#####
## PROCEDURE:   BEGIN
## PURPOSE:     Invoked once at the beginning of the run, to prime
##              the delivery lines.
## INPUTS:      $Col - Comma-separated list of active columns.

NEW BEGIN $Col <multiline>
  TRANSfer    Pressure(Amidite,Tet)      5
  TRANSfer    AToWaste                    0.2
  TRANSfer    GToWaste                    0.2
  TRANSfer    CToWaste                    0.2
  TRANSfer    TToWaste                    0.2
  TRANSfer    TetToWaste                   5
  TRANSfer    PressureCapAB                8
  TRANSfer    CapAToWaste                   5
  TRANSfer    CapBToWaste                   5
  TRANSfer    PressureIodine               5
  TRANSfer    IodineToWaste                5
  TRANSfer    PressureTCA                  15
  TRANSfer    TCAToWaste                    10
  TRANSfer    PressureDCM                  15
  TRANSfer    DCMToWaste                   10
  TRANSfer    PressureACN                  15
  TRANSfer    ACNToWaste                   10
  TRANSfer    ACNToColumn($Col)            10
  TRANSfer    ReverseFlush($Col)           10
  TRANSfer    BlockFlush                   10
  TRANSfer    BlockVent                    3
</multiline>

#####
## PROCEDURE:   DETRitylate
## PURPOSE:     Deprotection routine, invoked at every base addition
##              and optionally at the end of the synthesis run.
## INPUTS:      $Col - Comma-separated list of active columns.

NEW DETRitylate $Col <multiline>
  TRANSfer    BlockVent                    2
  TRANSfer    PressureTCA                  2
  TRANSfer    PressureACN                  2
  TRANSfer    PressureDCM                  2
  TRANSfer    DCMToWaste($Col)            25
  SAFE        No
  MONitor     TCAToWaste($Col)            110
  TRANSfer    FlushToWaste($Col)           7
  TRANSfer    ACNToWaste($Col)            12
  TRANSfer    FlushToWaste($Col)           7
  TRANSfer    ACNToWaste($Col)            12
  TRANSfer    ReverseFlush($Col)           7
  TRANSfer    ACNToColumn($Col)            12
  TRANSfer    ReverseFlush($Col)           9
  TRANSfer    ACNToWaste                   2
  TRANSfer    BlockFlush                   5
  SAFE        Yes
</multiline>

```

```

#####
## PROCEDURE: PREPare
## PURPOSE: Prepare for amidite delivery.
##           Invoked once per base addition.
## INPUTS:   $Col - Comma-separated list of active columns.

NEW PREPare $Col <multiline>
  TRANSfer BlockVent 2
  TRANSfer Pressure(Amidite,Tet) 3
</multiline>

#####
## PROCEDURE: DELIVer
## PURPOSE: Amidite delivery procedure. Invoked once for each
##           active column, at every base addition.
## INPUTS:   $Col - A single column
##           $Base - A single base to be delivered into the column
##           $TTime - Amidite delivery time (Set with TTIME)

NEW DELIVer $Col $Base $TTime <multiline>
  TRANSfer TetToColumn($Col) 1.7
  TRANSfer ($Base,Tet)ToColumn($Col) 5.0
  TRANSfer TetToColumn($Col) $TTime
  TRANSfer FlushToColumn($Col) 1
</multiline>

#####
## PROCEDURE: COUPlE
## PURPOSE: Couple procedure, Invoked once per base addition.
## INPUTS:   $Col - Comma-separated list of active columns.
##           $CTime - Coupling time (Set with CTIME)

NEW COUPlE $Col $CTime <multiline>
  SLEep $CTime
  TRANSfer ACNTowaste 4
  TRANSfer ReverseFlush($Col) 8
  TRANSfer BlockFlush 5
</multiline>

#####
## PROCEDURE: CAP
## PURPOSE: Cap synthesis columns. Invoked once per base addition.
## INPUTS:   $Col - Comma-separated list of active columns.

NEW CAP $Col <multiline>
  TRANSfer BlockVent 2
  TRANSfer PressureCapAB 2
  TRANSfer CapABToColumn($Col) 12
  SLEep 6
  TRANSfer ACNTowaste 4
  TRANSfer ReverseFlush($Col) 8
  TRANSfer BlockFlush 5
</multiline>

#####
## PROCEDURE: OXIDize
## PURPOSE: Oxidization routine, invoked once per base addition.
## INPUTS:   $Col - Comma-separated list of active columns.

NEW OXIDize $Col <multiline>
  TRANSfer BlockVent 2
  TRANSfer PressureIodine 2
  TRANSfer IodineToColumn($Col) 12
  TRANSfer BlockFlush 4
  SLEep 20
  TRANSfer ReverseFlush($Col) 8
  TRANSfer ACNTowaste 2
  TRANSfer ACNToColumn($Col) 13
  TRANSfer FlushToColumn($Col) 7
  TRANSfer ACNToColumn($Col) 13
  TRANSfer ReverseFlush($Col) 7
  TRANSfer ACNToColumn($Col) 15
  TRANSfer FlushToColumn($Col) 9
  TRANSfer BlockFlush 5
</multiline>

```