

Environmental Sample Processor: Protocol Parameters

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Core Protocol Methods

- **HAB, LARV, BAC, PUFM, KAR, DA, STX, and standAloneWCR**
 - All take two optional parameters, plus
 - an optional code block for non-standard clean up
- Ruby requires all default parameters to follow explicit ones
 - This means that one must specify the sample volume if options are specified

Specifying Sample Volumes

- First protocol method parameter, if specified, overrides default sample volumes
- A single non-negative Float value overrides only sh1 volume
- Last element of an array of 2 non-negative values overrides WCR volume
- 1st element overrides sh1 volume as in the single volume case
- Last element of nil causes WCR puck to be skipped
- First element of nil skips sh1 sampling (not puck)
- Sh1 volume must always be numeric. Sh1 cannot be skipped.
- Examples:
 - **hab 123** #123ml sh1 with default WCR
 - **larv [123, nil]** #123ml sh1 without WCR puck
 - **stx [123, 45]** #123ml sh1 with 45ml WCR
 - **bac 123, 45** # INVALID – 45 is not an option hash
 - **bac 123, nil** # INVALID – nil is not an option Hash
 - **da** #default da sample and WCR volumes
 - **hab nil** #negative lysate test

Default Sample Volumes

- Each protocol has a corresponding *\$protocolVol* global variable
- *\$protocolVol* is the default value of the protocol's first parameter
 - Examples:
 - *\$habVol=123* #default 123ml default HAB sh1 with default WCR
 - *\$habVol=[123, nil]* #123ml default HAB sh1 skip WCR pucks
 - *\$larvVol=[123, 45]* #123ml default LARV sh1 with 45ml WCR
 - *\$larvVol=123, 45* # VALID – same as [123,45] above
 - *\$habVol=123, nil* # VALID – default to skip HAB WCR pucks
- If *\$protocolVol* is a single numeric value, default WCR volume is determined by protocol's *:wcrVol* option
 - Discussed in upcoming “Protocol Options” slide
- If *\$protocolVol* is a two element array,
 - last element determines the WCR volume, overriding *:wcrVol* option

Protocol Options

- 2nd protocol method parameter, if specified, overrides default *\$protocol* options
- Each option is identified by a symbolic name – its hash key
- Ruby hashes merely map arbitrary keys to corresponding values
- **Hash** Examples:
 - `{ :a => 4, :b => 3.14, :foo => "foobar" }`
 - `{ 0 => 1, 1 => 0 }`
- Note that keys need not be Symbols, but they usually are.
- Curly braces must be omitted when passing Hash literal to methods
 - Otherwise, it would look like passing in a block of code!!
 - `p { :a => 4, :b => 3.14, :foo => "foobar" } #INVALID`
 - `p :a => 4, :b => 3.14, :foo => "foobar"`
 - `p a: 4, b: 3.14, foo: "foobar"`
- Undefined hash keys return nil
 - `{ :a => 4 }[:b] == nil`

Whole Cell Archive Options

- **:wcrVol** => default WCR sample volume or nil to skip WCR
- **:wcrFixes** => default list of wcr fixes
as in: **[:fix]*3** #3 fixes using the usual fixative
- **:wcrTime** => total WCR fix time
as in: **'60:00'** #each fix's duration is divided by **:wcrFixes.size**

Basic Protocol Options

- **:assay** => assay name (**:hab**, **:larv**, etc.)
Identifies the assay in the log and image file names
- **:sampler** => Sampler object to use (usually **\$protocol/Sampler**)
- **:image** => array of camera exposures
as in: **[5, 10, 40, :hires,160, :midres, :auto]**
- **:blockDelay** => time to delay for sh2 blocking
as in: **'8:00'** #8 minutes by default
- **:delay** => length of delay to plumb sample for short assays
- **:fast** => true or false #enables faster cleanup logic for lab use

Less Common Protocol Options

- **:email** => Email object specifying who should receive mail
 - As in: **Email.new ['brent','esp001'], :Subject=>"Progress"**
- **:samplePass** => procedure (code) to use for passing sample to PS
- **:delay** => length of delay to plumb sample for short assays
- **:flush** => flush procedure

Default Protocol Options

- Each PROTOCOL has a corresponding *\$protocol* global variable
- *\$protocol* is the default value of the protocol's second parameter
- It will always be a **Hash** mapping option names to values
- To change a default protocol option for all subsequent invocations:
\$protocol[:option] = newValue
- To override default protocol option(s) for only one invocation:
protocol [sh1Vol, wcrVol],
\$protocol.dup.update :option=>newValue
- First parameter must be specified in order to specify second one.
- **dup** prevents the default \$protocol options from being changed
 - By creating a copy of the options **Hash** and changing the copy

Example: HAB Protocol Options

-> \$hab

```
{assay => hab,  
email => HABfans,  
image => [hires, 160, default, 40, midres, auto],  
sampler => HABsampler,  
wcrSampler => HABWCRsampler,  
wcrVol => 50}
```

-> \$hab[:wcrVol] = 1500

1500

-> \$hab

```
{assay => hab,  
email => HABfans,  
image => [hires, 160, default, 40, midres, auto],  
sampler => HABsampler,  
wcrSampler => HABWCRsampler,  
wcrVol => 1500}
```

standAloneWCR Protocol Options

- There is no `$standAloneWCR` global variable
 - Because WCR is normally incorporated into other assays
- The WCR sample volume must be the 2nd specified.
- Best to define `myWCR` for your specific needs:

```
def myWCR vol=$myWCRvol, opts=$myWCRopts, &cleanup  
  standAloneWCR([nil, $vol], opts, &cleanup)  
end
```

- Setup:
`$myWCRvol = 100` #default sample volume
`$myWCRopts = $bac.dup.update wcrTime: "5:00"`
- Usage:
`myWCR` #defaults to 100ml
`myWCR 150` #sample 150ml

HABDA composite Protocol

- A single protocol combining HAB and DA assays
 - Harmful Algal Bloom
 - Including optional Whole Cell Archive
 - Demonic Acid
- Uses 6 or 7 pucks in all
- Parameters are:
 - HAB sample and WCR volumes
 - DA sample volume
 - Combined HAB and DA options
 - As the last **Hash** parameter

Sampler Objects and Options

- **Sampler** objects are created with `Sampler.new` as in: `defaultSampler = Sampler.new`
- The `Sampler.new` method takes a single optional parameter A **Hash** of sampler options!
- **Sampler** objects respond to named methods for each option as in: `defaultSampler.vacuum #outputs 10..13`
- Options in existing samplers may be changed as in: `defaultSampler.vacuum = 15..19 #to increase deltaP`
- Some Sampler object options are themselves complex Objects `defaultSampler.exhaleConfig` is
Servo configuration for exhalation

Sampler Options and default values

This **Hash** is returned by **Sampler.default**

:bubblePt => nil #bubble point of filter in psi
:vacuum=>10..13 #deltaP limits in psi while sampling
:maxDelay=>40 #max. seconds to delay to top of stroke
:minRate=>0.2 #min. rate before “clogged” in ml/second
:evacDelay=>15 #delay in seconds during puck evacuation
:numberOfEvacs=>1 #number of cycles of puck evacuation
:maxPumpVolume=>40 #max mls allowed to compress air to bubblePt
:equalized=>1.0 #call deltaP “equal” if abs(top-bottom)<= this in psi
:minPSI=>2.5 #minimum allowed pressure in syringe in psia
:maxResidue=>0.1 #minimum sample stroke volume in ml
:primeVolume=>7 #volume for initial priming sample stroke in ml
:exhaleConfig=>nil #default SS servo configuration for exhale stroke

- **Sampler.default[:bubblePt]=35** #change default bubble point to 35 psi

Protocol Sampler Objects

- Each core protocol defines its sampler **Object** as *\$protocol/Sampler*
- (only if no *\$protocol/Sampler* is already defined)

```
$habSampler ||= Sampler.new :bubblePt=>24
```

```
$larvSampler ||= Sampler.new :bubblePt=>24
```

```
$bacSampler ||= Sampler.new :numberOfEvacs=>2, :bubblePt=>70,  
                           :minRate=>25/(3*60.0), :vacuum=>20..24
```

```
$karSampler ||= Sampler.new(:bubblePt=>10, :vacuum=>4..7)
```

- Each of these is defined in the respective *PROTOCOL.rb* file
 - e.g. *BAC.rb*, *LARV.rb*, *HAB.rb*, or *KAR.rb*
- **:sampler** key in protocol options provides a means of overriding protocol's default sampler for specific invocations