



# Environmental Sample Processor: Protocol Parameters



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# Predefined 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
- Most protocols are configured in the Ruby script file:  
`mission/protocfg.rb`

# 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 (still loads sh1 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

- 2<sup>nd</sup> 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
    - VS.
    - p :a => 4, :b => 3.14, :foo => “foobar”
      - alternatively
      - p a: 4, b: 3.14, foo: “foobar”
  - Undefined hash keys return nil
    - { :a => 4}[:b] == nil



# 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.with :option=>newValue*
- First parameter must be specified in order to specify second one.

# 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]
- **:email** => Email topic list specifying who should receive mail
  - eg: HABfans
- **:fast** => true or false #enables faster cleanup for lab use

# 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`
- `:wcrSampler =>` sampler to use for WCR water sample

# Assay Processing Options

- **:hybTemp** => sandwich hybridization temperature
- **:diluentVol** => volume of diluent to add
  - if < 0, the specified -volume is added before lysis (pre-diluent)
  - if > 0, the specified volume is added after lysis (post-diluent)
- **:remix** => true or false (defaults to false)
  - enable remix for case where diluentVol < 0
- **:lyseHeat** => target lysis temperature
- **:lyseTime** => lysis duration
- **:blockDelay** => time to delay for sh2 blocking  
as in: **8.minutes** #8 minutes by default

# Less Common Protocol Options

- **:samplePass** => procedure (code) to use for passing sample to PS
- **:delay** => length of delay to plumb sample for short assays
- **:flush** => flush procedure

# 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 2<sup>nd</sup> 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.with 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