Are non-targeted effects important in radiation protection of the environment?

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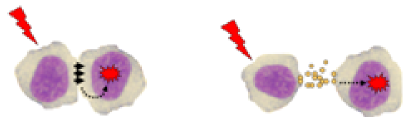
Who moved my cheese? - major underlying changes necessitating this discussion

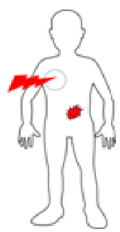
- Theoretical:
  - Shifting paradigms in radiobiology
- Practical:
  - Recognition that frequency of non-cancer diseases can be increased by radiation exposure
  - Recognition that non-human species deserve a specific protection framework

'Non-targeted' radiation effects

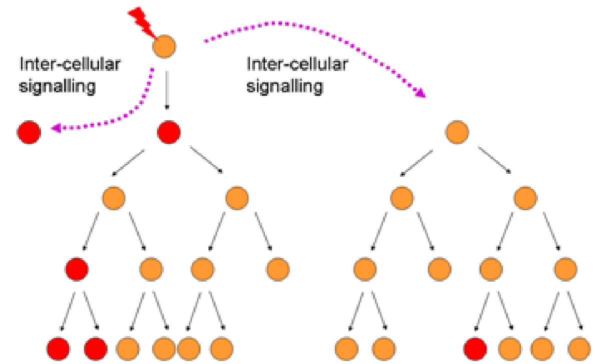
Bystander effects
Effects in neighbouring cells



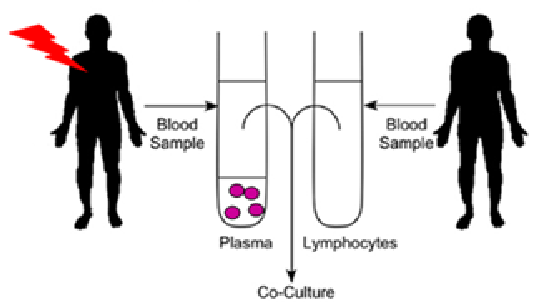
Abscopal effects
Effects in neighbouring tissues



Genomic Instability
Effects in unirradiated descendant cells



Clastogenic factors
Ex vivo effects in cultured cells



Inflammatory Processes may provide mechanistic link

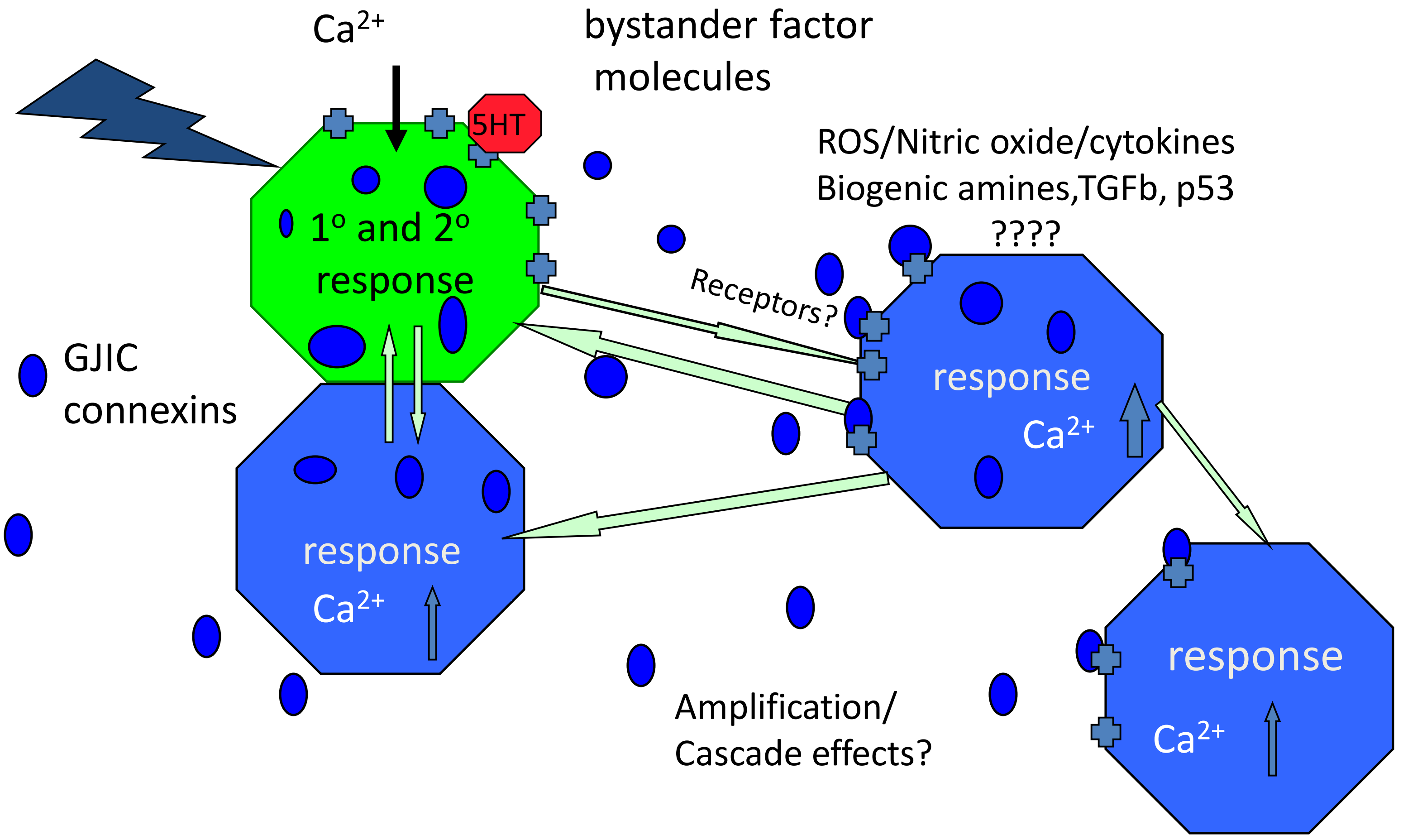
Inter-animal signaling

Effects in neighbouring animals

Long-term effects on innate immune
Response function may occur

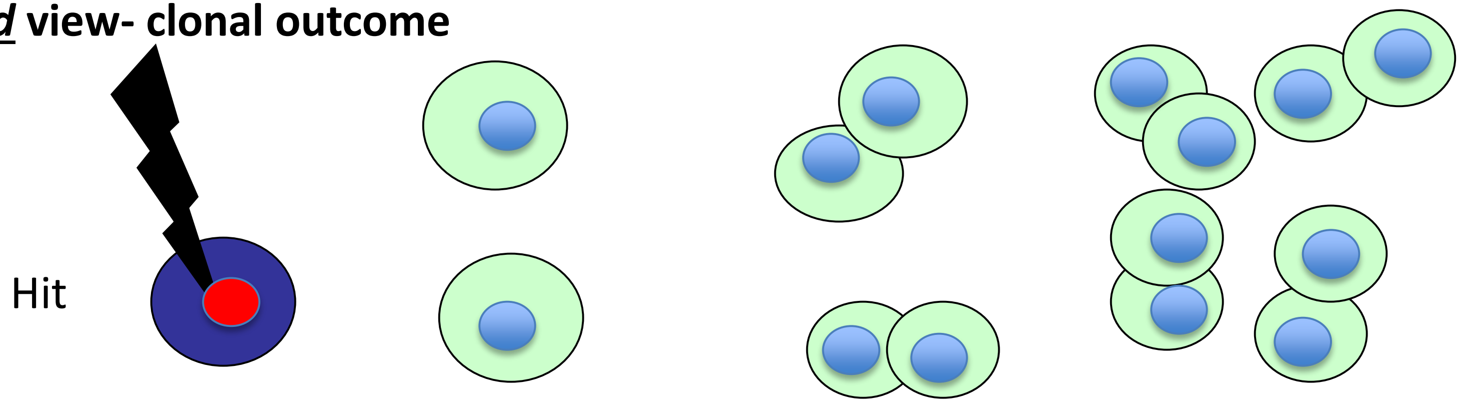
The bystander effect

Ionizing radiation, UVA, UVB, ELF-EMF and heavy metals induce affected cell to signal to others. Responses to the signals include apoptosis, micronucleus formation, transformation, mutation, induction of stress and adaptive pathways. Serotonin (5HT) and Calcium ions known to be involved in signal production.



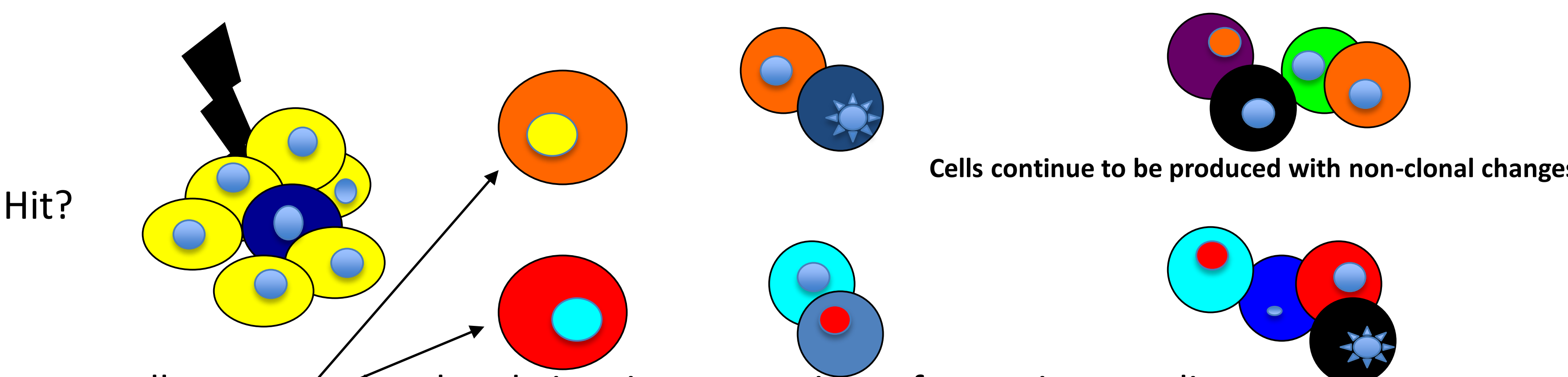
The link between bystander effects and genomic instability – twin pillars of the new paradigm

Old **view- clonal outcome**



Progeny are all clonal i.e. identical and mutation is passed to all progeny

New **view-non-clonal, population-determined outcome**



Progeny cells are non-clonal and give rise to a variety of mutations or die

Low dose effects are different

- Adaptive effects – not only strict radiobiological adaptive response but long-term evolutionary acclimation
  - Stuart and Boreham labs
- Hormetic effects – low dose of radiation is beneficial leading to non-linear dose responses for a variety of endpoints
  - Calabrese reviews, Boreham lab
- Homeostatic effects- systems accommodate and adjust to low dose induced perturbations
  - Seymour, Mothersill proteomics data, Tapio Lab
- Genetic and environmental factors more important than dose
  - Oughton/Salbu, Mosse/Marozik, Ullrich, Wright, many others

Chronic v Acute effects

- Not simply related – complicated by low dose responses such as adaptation already referred to
- Depend on assimilation which varies between species
- Depends on reproductive strategy and life cycle
- Depends on the isotope and its chemical function, speciation and abundance of competing elements

DDREF of 2 is simplistic

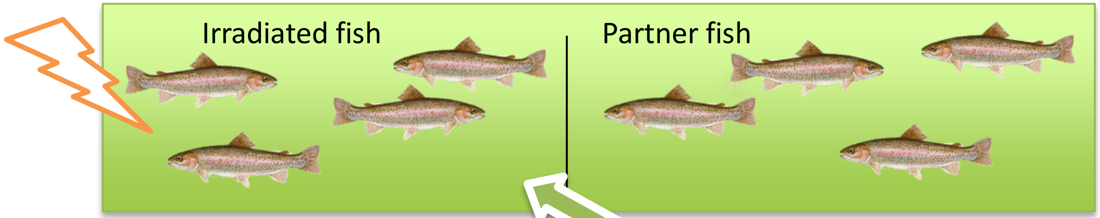
Current approaches

- Transfer factors and dose conversion factors based mainly on assumptions due to data gaps
- Modeling also based mainly on assumptions

Risk assessment tools such as
ERICA and ResRad Biota

BUT
What about reality?

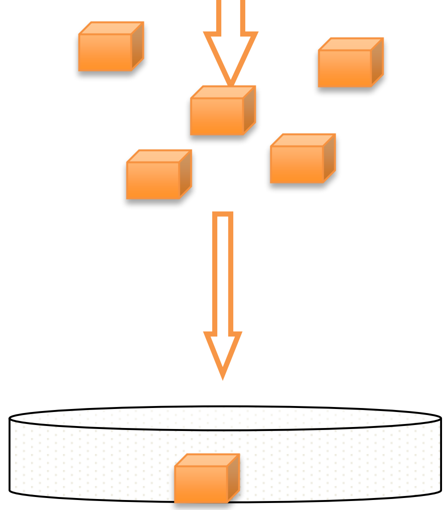
Measuring bystander response to radiation *in vivo* (adapted from Mothersill et al 2006)



Irradiate or sham irradiate fish, allow to swim with unexposed partner for 2hrs

Unexposed fish introduced into water from irradiated or sham fish After 2hrs. Dissect tissues

Do proteomics/histology



Explant pieces taken from skin, fin, gill, spleen and kidney

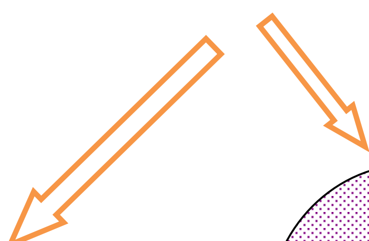
Do tissue culture

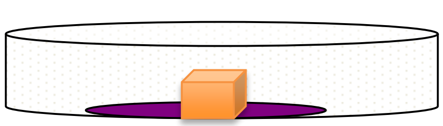
Culture of explants for 2 days

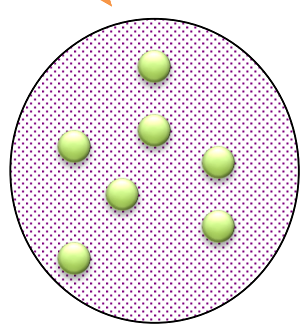
Harvest culture medium for calcium flux, ELISA and clonogenic assays

Grow up culture

examine explant outgrowth do immunocytochemistry





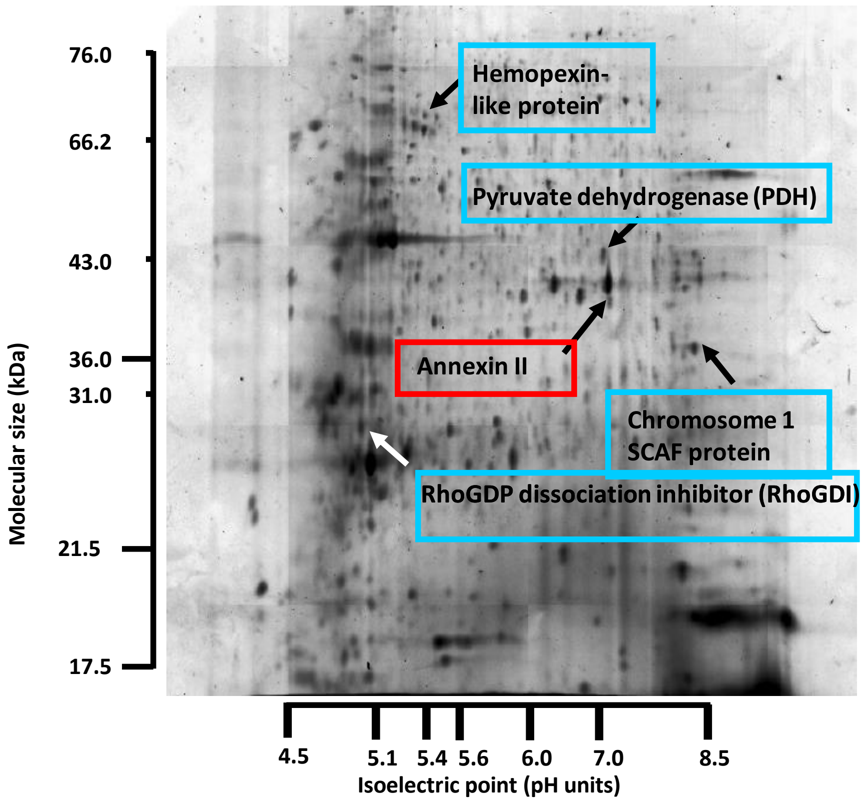


Add medium to unirradiated clonogenic cell line

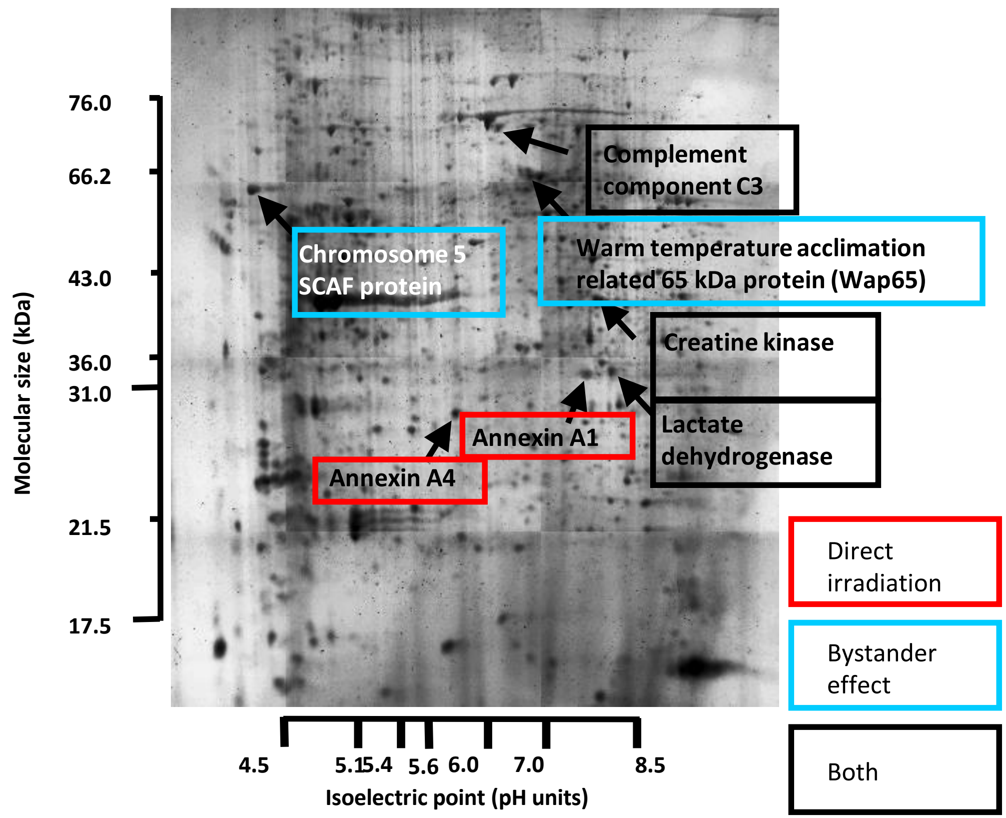
determine surviving fraction by counting colonies after 10 days

Gill proteomics in two species

Rainbow trout



Medaka



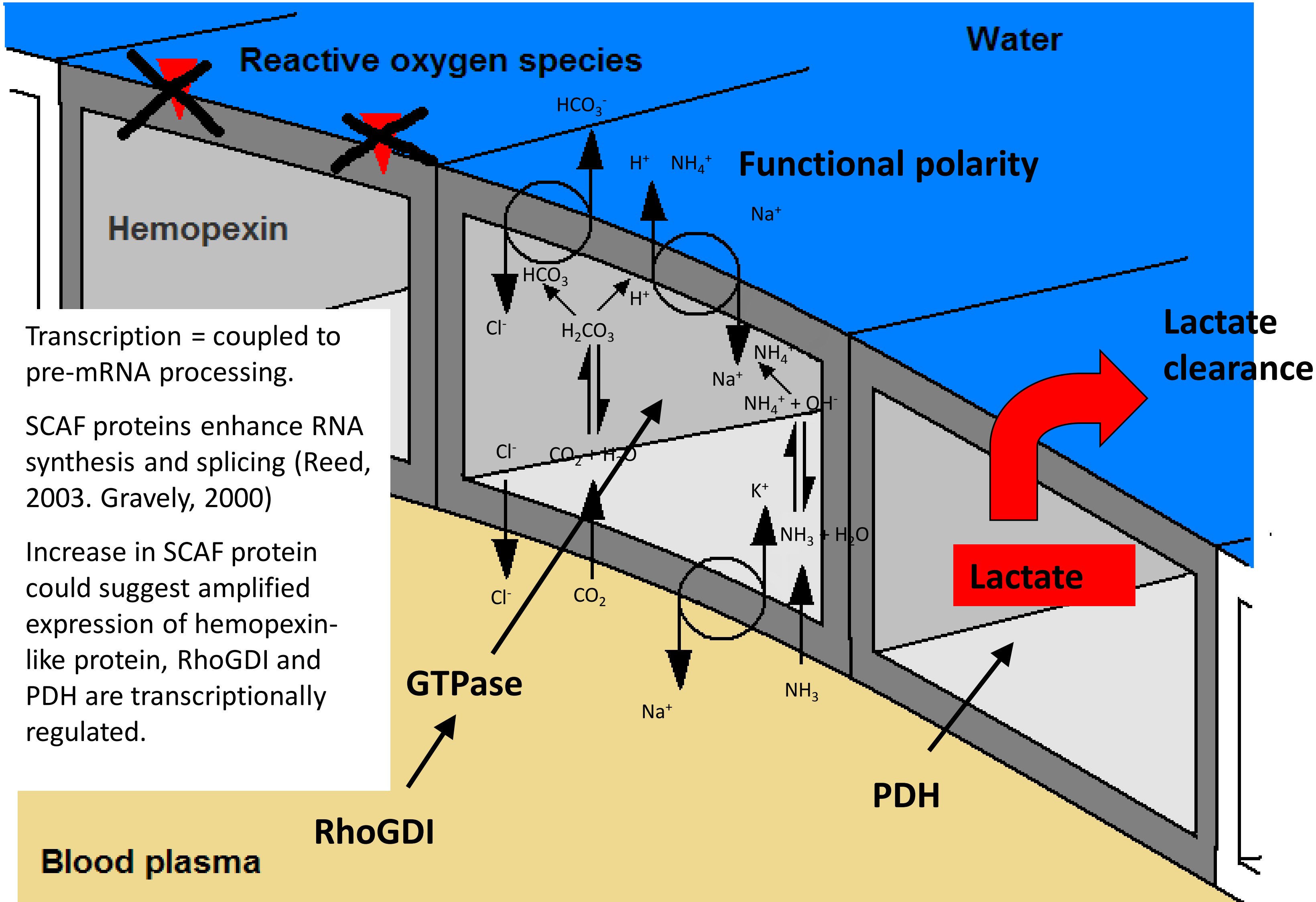
Trout bystander proteome protective

Smith RW, et al 2007 Evidence for a protective bystander response in rainbow trout gills exposed to x-irradiation. Proteomics. 7(22):4171-80.

Proteomic changes in the gills of DNA repair proficient and DNA repair deficient medaka following exposure to direct irradiation and to X-ray induced bystander signals. R Smith et al BBA being revised after review

Medaka bystander proteome may indicate protective and adaptive response

INTEGRATIVE PROTECTIVE RESPONSE TO THE BYSTANDER SIGNAL IN THE TROUT GILL

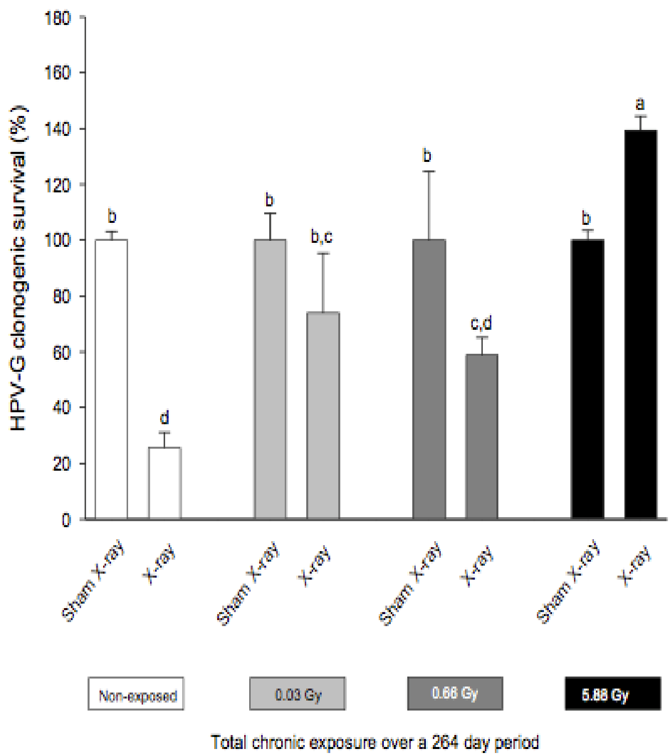


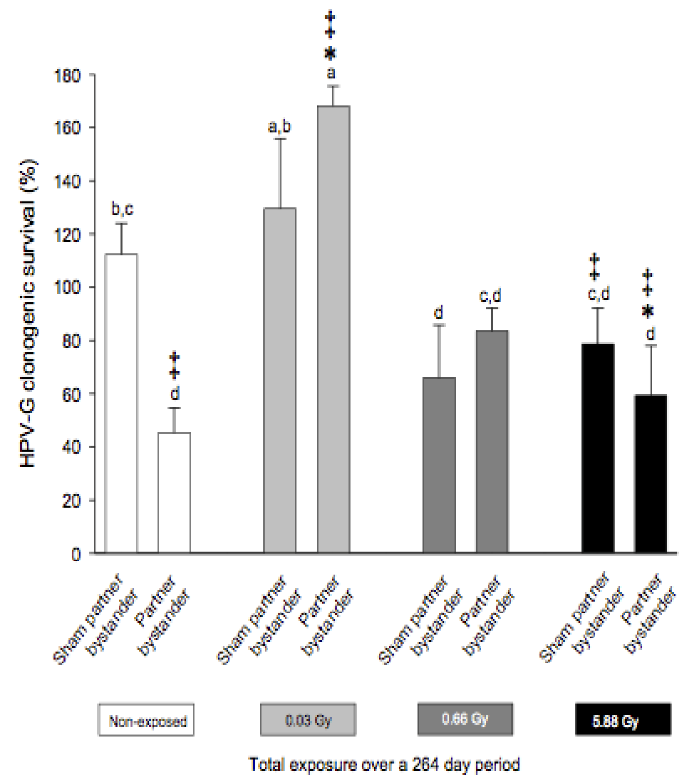
Transcription = coupled to pre-mRNA processing.

SCAF proteins enhance RNA synthesis and splicing (Reed, 2003. Gravely, 2000)

Increase in SCAF protein could suggest amplified expression of hemopexin-like protein, RhoGDI and PDH are transcriptionally regulated.

Chronic Medaka low LET data suggests protective/adaptive responses

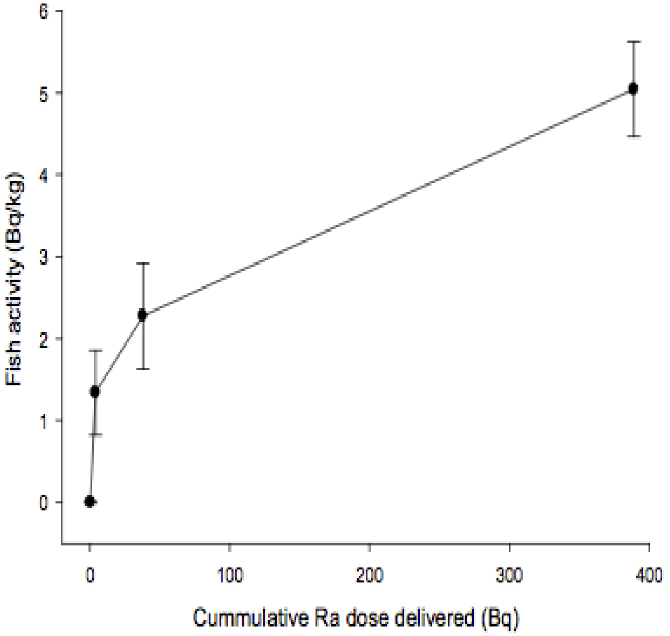




Example of chronic high LET data

ID	Activity ($Bq\ kg^{-1}$ wet)	Annual dose ($mGy\ y^{-1}$)
Control Fish	36 ± 22	0,9 ± 0,5
Control Fish	28 ± 28	0,7 ± 0,7
Fed 10 $mBq\ g^{-1}$	39 ± 15	1,0 ± 0,7
Fed 10 $mBq\ g^{-1}$	23 ± 8	0,6 ± 0,2
Fed 100 $mBq\ g^{-1}$	11 ± 12	0,2 ± 0,2
Fed 100 $mBq\ g^{-1}$	9 ± 12	0,2 ± 0,3
Fed 1 $Bq\ g^{-1}$	26 ± 11	0,7 ± 0,3
Fed 1 $Bq\ g^{-1}$	33 ± 13	0,8 ± 0,3
Fed 10 $Bq\ g^{-1}$	100 ± 18	2,5 ± 0,4
Fed 10 $Bq\ g^{-1}$	124 ± 16	3,0 ± 0,4

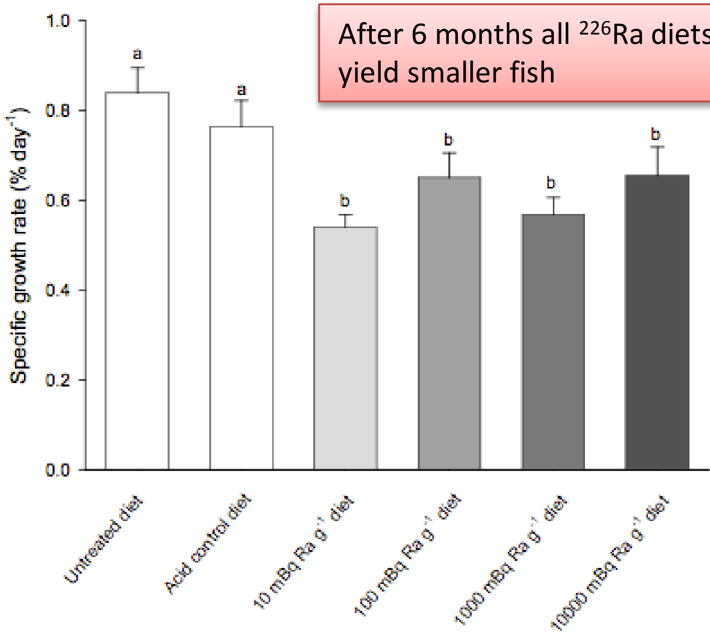
Approx 50μBq per Bq assimilated in 6 months



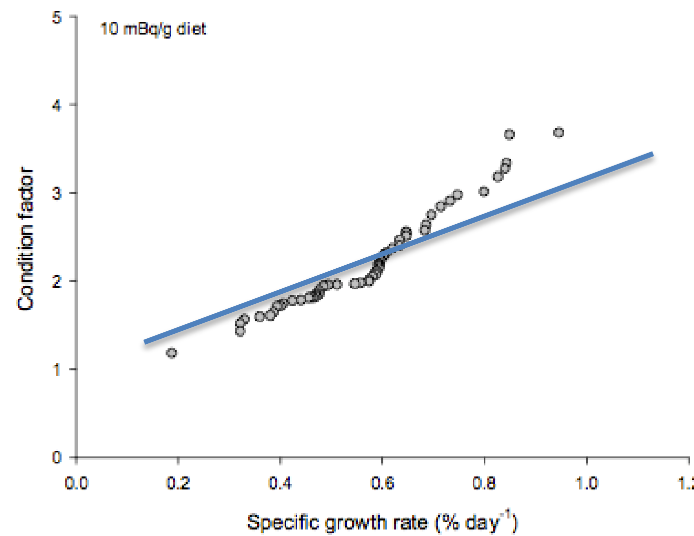
Despite very low retention biological *effects* ARE seen

Relationship between K and SGR deviates in Ra fed fish. Points above the line show small (slow growing) fish with greater than expected K factor
SMALL FAT FISH!

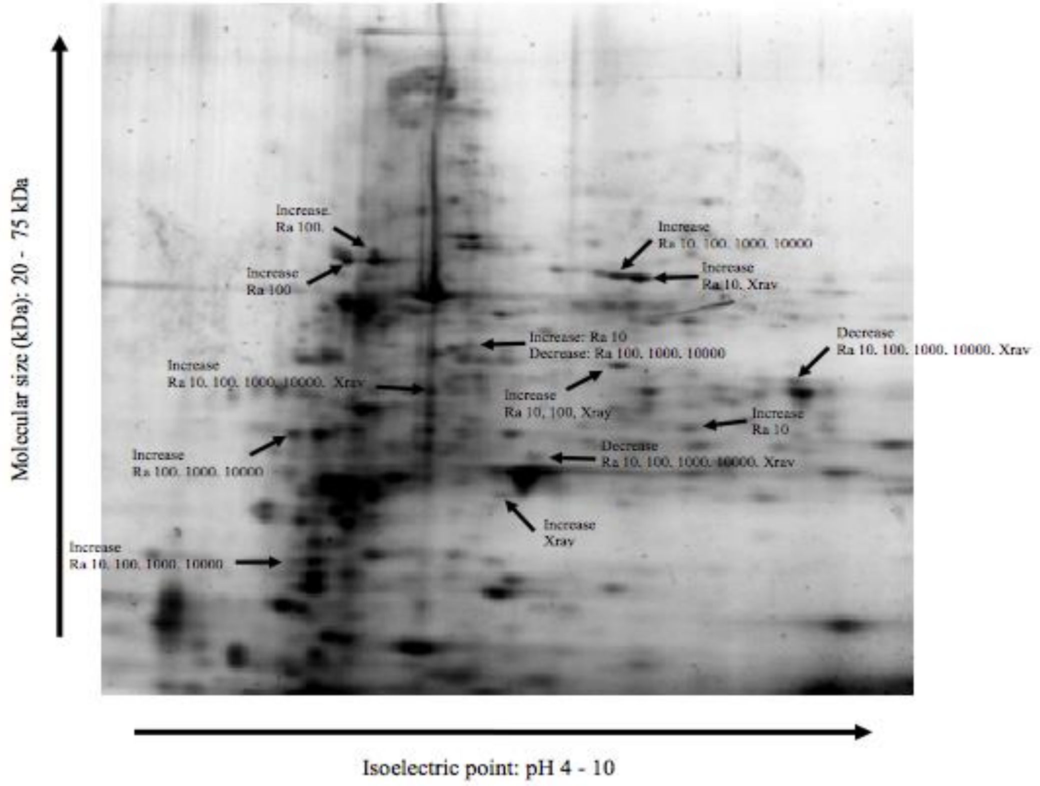
After 6 months all ^{226}Ra diets yield smaller fish



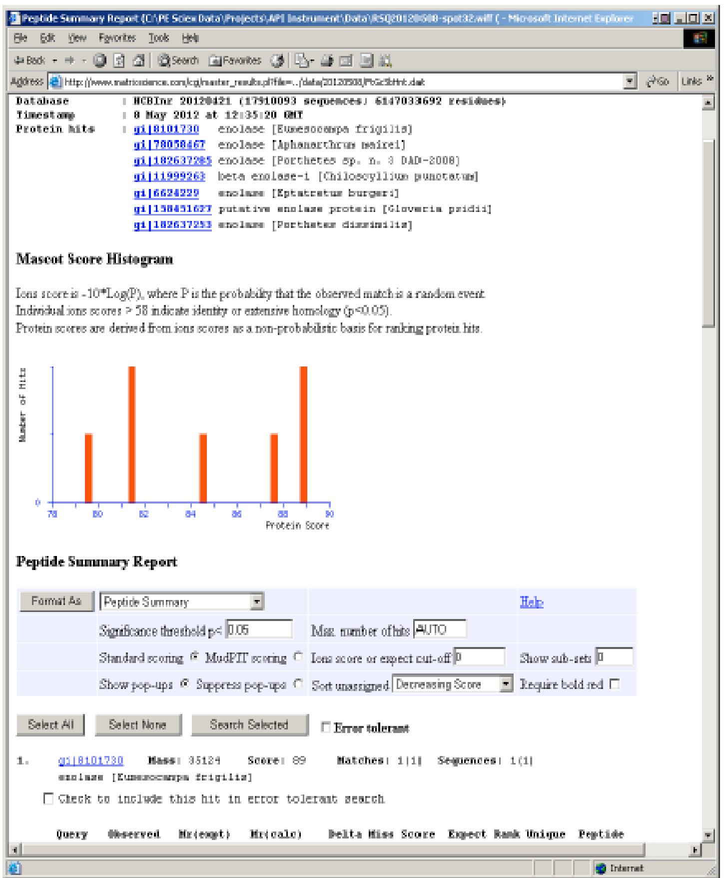
1 year on diet (10mBq/g)

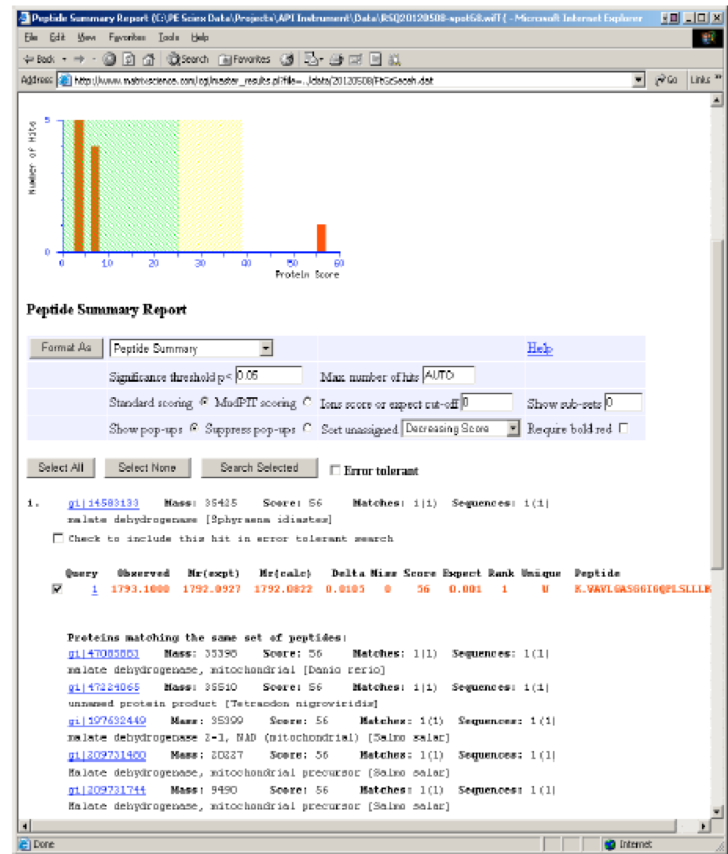


Preliminary proteomics data after 6 months (where growth anomalies occur)

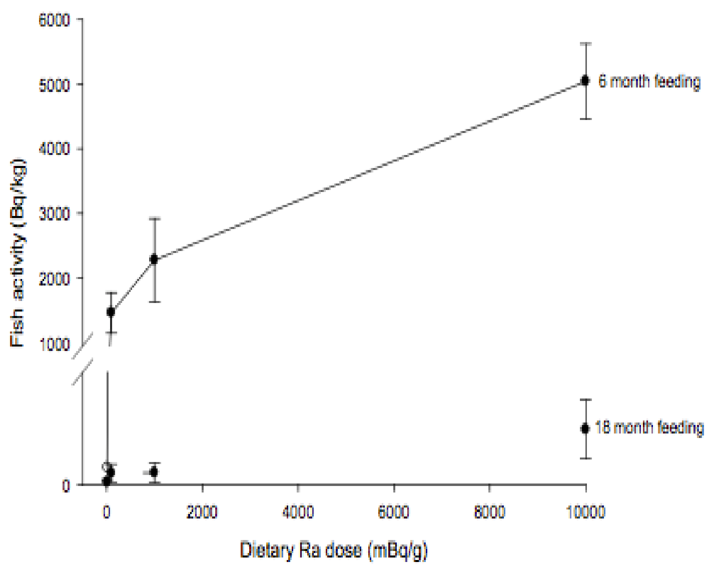


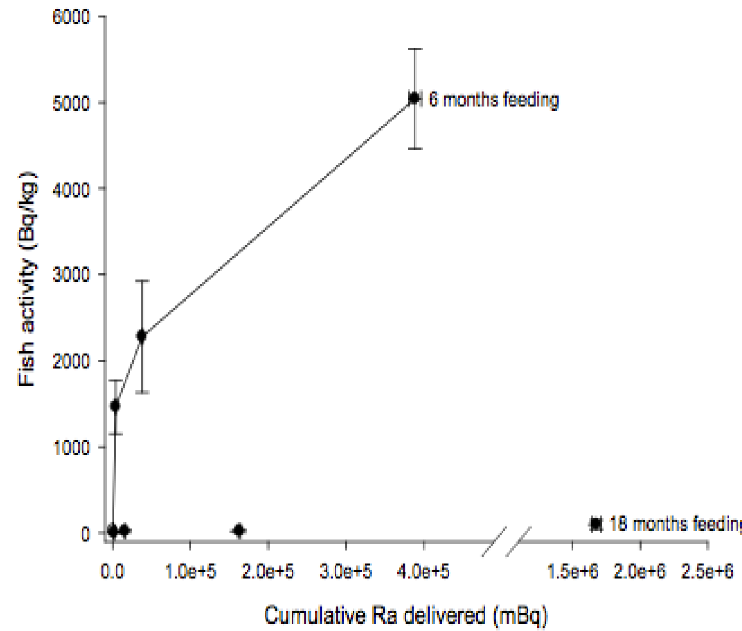
Initial protein identities: Gel spot 32 Enolase, increased by 10 mBq/g only and a 0.5Gy X-ray and spot 68 malate dehydrogenase, decreased by all Ra diets and X-ray





Comparison of 6 and 18 months showing loss of accumulated Ra-226 at 18 months





Summary points to consider

- Horizontal and vertical transmission mean the “target” is not confined to the cell or organism receiving the dose
- Need to consider the hierarchical level at which damage (effect/response) is being assessed or is of concern
- May need to define new critical endpoints including emergent properties
- Need to be careful about interpretation of effects data at levels lower than the individual organism

Acknowledgements

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Thank you!