Radioactive protein-labelling techniques

SIR-We wish to alert the scientific community to the existence of a problem which, though widespread, has heretofore gone largely unreported. The use of ⁸S-labelled amino acids (methionine, cysteine) to label proteins for further study is an almost universal practice in laboratories conducting molecular and cell biology. We assume that the precautions to be taken when using 35S, a lowenergy β -particle emitter, are generally recognized. Generally unappreciated, however, is the fact that solutions of ³⁵Slabelled amino acids release a volatile radioactive component which can pose a containment problem. It is released from every batch of ³⁵S-amino acids we have tested, regardless of the manufacturer or specific amino acid (though there is some suggestion that less is released from ³⁵S-cysteine than from ³⁵S-methionine).

When and where is this radioactive component most likely to be encountered? When a fresh vial of 8 mCi ³⁵Samino acids is thawed, without a lid, in a large, closed container, approximately 1 µCi ³⁵S, as determined by standard airsampling techniques, is released into the air. This may result from product breakdown occurring during the freezing process (which is known to accelerate physicochemical breakdown). Further volatile material is generated when ³⁵S-amino acids are added to cell culture medium and incubated at 37 °C, regardless of whether cells are present; thus this component is not generated metabolically. If it is indeed the result of a chemical/physical breakdown, the addition of stabilizers to the amino acids should decrease the amount of this component produced.

The volatile component is very soluble in water; thus the water present in incubators used for cell culture can become contaminated. A total of 300,000 c.p.m. (measured by liquid scintillation counting) were found in 500 ml water consequent to the use of 2.5 mCi ³⁵S-methionine in a single, 6-h incubation. Because this water is continually evaporating, recondensing and running down the inside of the incubator door, all the surfaces in these incubators - trays, side walls, door and even the outside of other dishes of cells-may become contaminated. The rubber gasket sealing the door and the metal fan which recirculates the air inside the incubators were found to be so highly contaminated that this is readily detectable by a hand-held G-M monitor. Filters used to wipe 10-cm² areas inside the incubator picked up several thousand c.p.m.

Although it is embarrassing that few researchers have picked up this problem directly, it is also disturbing that the manufacturers of ³⁵S-amino acids do not include more specific warning in their product-information sheets. All three

main producers are aware of the potential for such a problem, yet none, until very recently, has found time to investigate it.

-SCIENTIFIC CORRESPONDENCE-

While we await the manufacturers' recommendations, there are several precautions that are easily taken and which we find to be very effective in limiting contamination. First, vials of ³⁵S-amino acids should be thawed in a fume hood using a needle through the rubber septum to vent the vial. Alternatively, and perhaps more effectively, a syringe packed with charcoal (similar to those used for ¹²⁵I) attached to such a needle could be used.

Second, as activated charcoal readily absorbs the volatile radioactivity, we have placed a 16×16 inch pressed-charcoal filter of a honeycomb-type design on the top shelf of our incubator. This filter does not affect the CO₂ equilibrium in the incubator and, judging by the routinely low c.p.m. of air samples but increasing c.p.m. absorbed in the filter, it effectively

decreases contamination in the air. For less frequent use, activated charcoal in a tray or wrapped in several tissues to make a small bag will decrease contamination of the incubator.

Third, the water inside incubators should be changed on a regular basis, ideally after each labelling.

What is the volatile radioactive component? Two likely candidates have been named by the manufacturers: SO_2 or CH₃SH. At least two companies are working on its identity, and on solutions to the contamination problem. We expect a more detailed description of both the volatile radioactive component and the hazards it may pose on product-specification sheets in the very near future.

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Taxonomic instability continues to irritate

SIR—In his article on taxonomic instability, D. L. Hawksworth¹ may have aimed at the wrong target. We agree that name changes resulting from antiquarian research are usually unnecessary and that little would be lost if they were eliminated. But in our experience by far the most tiresome changes arise from what Hawksworth charitably calls "advancement of scientific knowledge", but which are often better described as mere changes of opinion as to where or whether, in an assembly of species, a generic distinction should be placed.

Systemics has been bedevilled by a failure to agree on the purpose of taxonomy. To Linnaeus it was simply to give a name to a species, just like naming an individual in a human family. With the acceptance of evolutionary change came the hope that taxonomy might reflect phylogeny. Few realized what a Pandora's box was being opened up. For the two functions could be acceptably combined only if everyone held identical views not only on the genealogy of plant and animal life, but also of the closeness of the relationship between species of a genus.

The conception of the genus uniquely displays the inevitable ambiguity: on the one hand, like family, class and highertaxa, it is a noun without objective reality. It is only an abstract view, based on an opinion, of which characters are of greater or lesser importance in phylogeny and therefore of where taxonomic divisions should be placed. By contrast, the species is a noun representing a real entity, at least as far as sexually reproducing organisms are concerned. But, unlike all other higher taxa, the genus also forms part of the name of the species. Thus, if a species is thought to be sufficiently misplaced among its fellows, its position cannot be remedied without changing its name, thereby messing up past literature, confusing the language with different nouns for the same entity and imposing additional burdens on the memory.

It is the generation of genera that requires suppression, even more urgently than the suppression of priority searchers. Whatever established taxonomists say, unless this ever-increasing problem can be dealt with, the volatility of generic names will continue unabated until every species is allocated to its own genus.

Fortunately the remedy is simple, but it will negate one of the current recommendations of taxonomic legislators. It is to promote a half-way house between the genus and the species, namely the despised subgenus, which, like all other taxa beyond the species, would not be part of the species name. This would enable species names, such as the present binomials, to remain immutable, as logically all nouns should be in scientific literature, while allowing taxonomists the freedom to move species into or out of whichever subgenera are available or which they think need to be created.

As an example of the usefulness of subgenera, one may cite Darwin's work² on six-plated barnacles comprising the genus *Balanus*. He realized, of course, that narrower groupings were possible and accordingly divided the genus into sections. Pilsbury named these sections as subgenera, yet the literature remained intact³. But Newman and Ross⁴, though adding little new to knowledge, renamed Pils-