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Barrett A J & Starkey P M. The interaction of α_2-macroglobulin with proteinases: characteristics and specificity of the reaction, and a hypothesis concerning its molecular mechanism. *Biochemical J.* 133:709-24, 1973.
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We showed that the plasma protein α_2-macroglobulin binds active proteinases almost regardless of their specificity or catalytic mechanism. We proposed a model for the interaction in which the proteinase cleaves a peptide bond in a sensitive region of the macroglobulin, and this results in a conformational change that traps the enzyme irreversibly. The active site of the trapped proteinase is sterically hindered, so that small substrates and inhibitors can interact with it, but not large ones. Physiologically, the conformational change could explain the rapid clearance of complexes from the circulation. [The *SCI*® indicates that this paper has been cited in over 555 publications.]

A Trap for Proteinases

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Our group had been making much use of antisera to lysosomal cathepsin D in our study of the mechanisms of cartilage breakdown in arthritis.[1] Phyllis M. Starkey and I were then trying to obtain antiserum to human cathepsin B. We expected that the antisera would inhibit the enzyme but were surprised to find that even nonimmune sera interacted with the enzyme in a way that resembled immunoinhibition. That is to say, the enzyme formed a higher molecular weight complex that retained some activity against synthetic substrates. We identified the serum component that bound the cathepsin in this way as α_2-macroglobulin (α_2M).[2] When we read what had been discovered by others about the interaction of α_2M with proteinases, we found its characteristics fascinating and initially inexplicable. The macroglobulin was able to recognize and bind a very wide range of proteolytically active molecules but also to distinguish them from very similar molecules that lacked proteolytic activity. We could show that all the proteinase molecules were bound in the same way, because they competed with each other, and when bound they showed the characteristics of steric inhibition that we had become familiar with in our work with antibodies.

We puzzled over the problem for some time, and then came the day when we were talking it over in my office, and the idea burst on us—that the active proteinase might initiate the interaction by cleaving a sensitive part of the macroglobulin molecule and that this could trigger a conformational change that resulted in the physical trapping of the proteinase molecule within it. The trapped proteinase molecule, being surrounded by the macroglobulin, would be shielded from contact with other large molecules, but small ones would be able to enter. So, this was the "trap hypothesis," and the part of the macroglobulin that was sensitive to proteolysis was the "bait region." This kind of protein-protein interaction was quite unlike any that was known, and we were initially very hesitant about the idea. We had no direct evidence for the proteolytic cleavage we were postulating, or for the major conformational change, but we gathered together a lot of circumstantial evidence that was consistent with our idea and submitted it to the *Biochemical Journal*. The editors were good enough not to make too many difficulties over either the rather speculative content of the paper or its considerable length.

We were not surprised that the trap hypothesis was met at first with a large measure of scepticism. We had no difficulty in agreeing that it seemed a most improbable kind of interaction, but the fact was that neither we nor anyone else had managed to come up with anything more plausible. Then, almost immediately, the kind of proteolytic cleavage we had envisaged was demonstrated by Peter Harpel, and we ourselves soon confirmed and extended his finding. Working with Ed Munn, we also obtained electron-microscopic evidence of the conformational change.[3] From then on, Phyllis and I were fairly confident that we were more or less right, but there were surprises in store. We had estimated the trapping capacity of the α_2M molecule as one molecule of proteinase, but others showed that it can normally bind two. Much more surprising, α_2M was shown to bind proteinases also by a covalent reaction, again triggered by bait region cleavage, but this proved to be less significant than the trapping, under normal circumstances.[4]

I think it is now generally accepted that the trap hypothesis is likely to be right in essence, and much new work on α_2M is interpreted in terms of it.[5,6]

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3. ————. α_2-Macroglobulin: a physiological regulator of proteinase activity. (Barrett A J, ed.) *Proteinases in mammalian cells and tissues.* Amsterdam, The Netherlands: North-Holland, 1977. p. 663-96. (Cited 190 times.)
4. **Salvesen G S, Sayers C A & Barrett A J.** Further characterization of the covalent linking reaction of α_2-macroglobulin. *Biochemical J.* 195:453-61, 1981. (Cited 110 times.)
5. **Feldman S R, Gonias S L & Pizzo S V.** Model of α_2-macroglobulin structure and function. *Proc. Nat. Acad. Sci. USA* 82:5700-4, 1985. (Cited 40 times.)
6. **Crews B C, James M W, Beth A H, Gettins P & Cunningham L W.** In support of the trap hypothesis. Chymotrypsin is not rigidly held in its complex with human α_2-macroglobulin. *Biochemistry—USA* 26:5963-7, 1987.

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