This Week's Citation Classic™
CC/NUMBER 29
JULY 16, 1984

Lindell T J, Weinberg F, Morris P W, Roeder R G & Rutter W J. Specific inhibition of nuclear RNA polymerase II by α-amanitin. *Science* 170:447-9, 1970.
[Dept. Biochemistry and Biophysics, Univ. California, San Francisco, CA]

α-Amanitin, the bicyclic octapeptide from *Amanita phalloides*, was found to be a potent and specific inhibitor of DNA-dependent RNA polymerase II from phylogenetically divergent organisms (rat and sea urchin). The utility of this inhibition was also demonstrated by differentially assaying RNA polymerase activities in isolated nuclei. [The *SCI*® indicates that this paper has been cited in over 450 publications since 1970.]

Thomas J. Lindell
Departments of Pharmacology
and
Molecular and Cellular Biology
University of Arizona
Tucson, AZ 85721

June 8, 1984

"I originally went to Bill Rutter's laboratory in fall 1968 on a postdoctoral fellowship to work on the comparative biochemistry of aldolases. Shortly after my arrival in Seattle, Bob Roeder began to obtain some striking results indicating that eukaryotes contained three separate RNA polymerases (I, II, and III).[1] Because of this excitement and the fact that I shared a laboratory with Bob, I chose to work on the RNA polymerase problem. The actual impetus for us to investigate the potential role of α-amanitin as an inhibitor of eukaryotic transcription was derived from a report by Stirpe and Fiume.[2]

"Rutter had obtained a sample of the toxin by telephoning T. Wieland[3] in Germany but Roeder did not have time to examine the effect of the compound on the individual RNA polymerases he had separated despite active discussions in the lab about the project. When Rutter moved from Seattle to San Francisco, I was the only person in the laboratory with experience who had actually separated and assayed the multiple RNA polymerases. The problem then became mine to determine whether α-amanitin inhibited any of these individual RNA polymerases.

"Fanyela Weinberg, who was a technician, and I did the original experiments with the sea urchin enzymes and Paul Morris repeated them on the separated enzymes from rat liver. As we were preparing a manuscript to submit to *Nature*, Pierre Chambon wrote to Rutter and enclosed a preprint on similar work describing an effect of α-amanitin on RNA polymerase B(II) which would be published shortly.[4] I was personally decimated by that communication, but because we had some different observations, we immediately submitted the paper to *Nature*, where it was rejected. Rutter was able to convince the editor of *Nature* to reconsider the paper but it was again rejected. Another paper describing the effect of α-amanitin on eukaryotic RNA polymerase by Jacob *et al.*[5] was published shortly thereafter in *Nature* and ours was finally published in *Science* in October 1970.

"The probable reason our paper was so highly cited was that ours was a more complete and comprehensive study and the results could be more easily extended to other work. Our paper also graphically demonstrated that RNA polymerase II from two highly divergent species was specifically inhibited after separation on DEAE Sephadex columns and that chain elongation was inhibited. Further, it demonstrated that a differential assay of the eukaryotic RNA polymerases could be obtained in isolated nuclei without resorting to their separation by column chromatography. Later, it was observed that RNA polymerase III was also inhibited by α-amanitin, but at higher concentrations.[6] Thus, using two different concentrations of α-amanitin, it is now possible to assay all three RNA polymerase activities in isolated nuclei."[7]

1. **Roeder R G & Rutter W J.** Multiple forms of DNA-dependent RNA polymerase in eukaryotic organisms. *Nature* 224:234-7, 1969. (Cited 670 times.)
2. **Stirpe F & Fiume L.** Studies on the pathogenesis of liver necrosis by α-amanitin. Effect of α-amanitin on ribonucleic acid synthesis and on ribonucleic acid polymerase in mouse liver nuclei. *Biochemical J.* 105:779-82, 1967. (Cited 160 times.)
3. **Wieland T.** Poisonous principles of mushrooms of the genus *Amanita*. *Science* 159:946-52, 1968. (Cited 230 times.)
4. **Kedinger C, Gniazdowski M, Mandel J L, Jr., Gissinger F & Chambon P.** α-Amanitin: a specific inhibitor of one of two DNA-dependent RNA polymerase activities from calf thymus. *Biochem. Biophys. Res. Commun.* 38:165-71, 1970. (Cited 370 times.)
5. **Jacob S T, Sajdel E M & Munro H N.** Specific action of α-amanitin on mammalian RNA polymerase protein. *Nature* 225:60-2, 1970. (Cited 95 times.)
6. **Weinmann R & Roeder R G.** Role of DNA-dependent RNA polymerase III in transcription of the tRNA and 5S RNA genes. *Proc. Nat. Acad. Sci. US* 71:1790-4, 1974. (Cited 345 times.)
7. **Lindell T J & Duffy J J.** Enhanced transcription by RNA polymerases II and III after inhibition of protein synthesis. *J. Biol. Chem.* 254:1454-6. 1979.