

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:	)	
	)	
Stephan HOLZSCHUH, et al.	)	
	)	
Serial No.: 17/295,647	)	
	)	
§ 371 Date: 20 May 2021	)	FORMULATION OF CONTRAST
	)	MEDIA AND PROCESS OF
Art Unit: 1618	)	PREPARATION THEREOF
	)	
Patent Examiner: James W. Rogers	)	
	)	
Confirmation No.: 2326	)	
	)	
Our Ref: BHC183030 PCT-US	)	

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION OF THOMAS FRENZEL

Dear Sir:

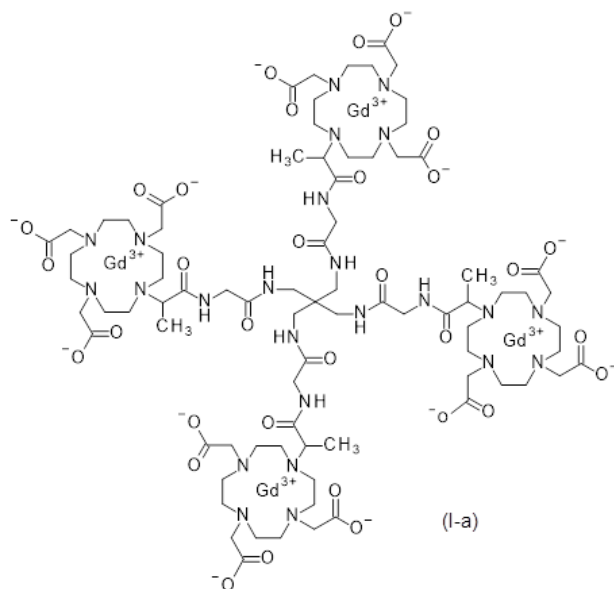
- 1) I am Thomas Frenzel, the undersigned, and I depose and declare as follows:

- 2) I received my State Examination in Pharmacy from the Ludwig-Maximilians University in Munich (Germany) in 1982. In 1987 I received my PhD in Pharmaceutical Biology from the Ludwig-Maximilians University in Munich (Germany) and performed Post Doctoral Studies at the Ludwig-Maximilians University in Munich (Germany) and at the University of Washington in Seattle (USA) (1987-1989).

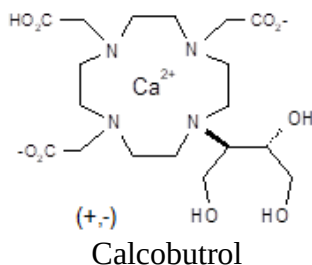
- 3) I am currently a Consultant at Bayer AG in Berlin, Germany in Pharmaceuticals, Radiology, MR and CT Contrast Media after being employed as a Senior Scientist at Bayer AG (previously Schering AG) in Berlin, Germany from 1989 to 2022. While at Bayer AG / Schering AG, I worked in the Department of MRI and X-Ray Contrast Media Research for 33 years. I am skilled in the art of gadolinium-based MRI contrast agents and pharmaceutical formulations thereof.

4) I am an inventor of the above-identified application, U.S. Patent Application Serial No. 17/295,647 (the '647 application), entitled Formulation of Contrast Media and Process of Preparation Thereof, and am familiar with the contents thereof.

5) In the claims of the '647 application, a liquid pharmaceutical formulation of a novel MRI contrast agent having a DO3A-derived tetra-chelate structure, described by Formula I in the '647 application, for example as shown in Formula I-a, below, in a pharmaceutically acceptable solvent is recited. The structure of the contrast agent has four Gadolinium ions (Gd^{3+}), each one chelated within a DO3A-derived ligand substructure. The DO3A-derived ligand structure is attached by a glycine tether to a central tetraamine core.



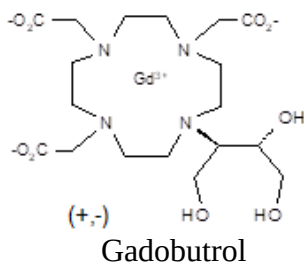
6) The liquid pharmaceutical formulation claimed in the '647 application further includes a small concentration of Calcobutrol (0.002% to 5% mol/mol Calcobutrol measured in proportion relative to the Gd concentration in the formulation), a compound capable of forming a chelate with free Gd^{3+} ions. Calcobutrol (structure below) has a structure of the Butrol ligand with a Ca^{2+} ion chelated therein.



7) The inventors of the '647 application have discovered that small amounts of Calcobutrol added to a formulation comprising a DO3A-derived tetra-chelate described in the

'647 application ensures the absence of free paramagnetic metal ions, such as Gd^{3+} , in the injectable formulation while preserving the performance levels as a contrast medium in medical imaging procedures (see, the '647 application as filed at page 3, lines 21-27).

8) During prosecution at the USPTO of the '647 application, it is my understanding that the examiner has rejected the pending claims as lacking inventive step (i.e., being obvious) in view of the combination of PCT International Application Publication No. WO 2016/193190 to Berger et al. (hereinafter "Berger") in view of German Patent Document DE102009053171 to Platzek et al. (hereinafter "Platzek," equivalent to PCT International Application Publication No. WO 2011/054827). I am familiar with and a listed inventor on Berger and am familiar with the disclosure and teachings of Platzek. Berger describes MRI contrast agents having a DO3A-derived tetra-chelate structure as shown in Formulas I and I-a in the '647 application. Platzek describes the formulation of the commercially available MRI contrast agent Gadavist which includes Gadobutrol (structure below) with small amounts of Calcobutrol as a metal scavenging agent. As shown in the structures, Gadobutrol and Calcobutrol have the same Butrol cyclic ligand structure and differ only in the identity of the complexed metal ion, i.e., Gd^{3+} for Gadobutrol and Ca^{2+} for Calcobutrol.



9) The addition of a metal scavenger to the formulation of a gadolinium-based contrast agent is desired in the art for two reasons. The first reason, as outlined below, is the recomplexation of Gd^{3+} ions which may be released from the gadolinium-based contrast agent during storage and shipping of the formulation. Even if the gadolinium-based contrast agent is of highest complex stability, as in the case of those described by Formula I in the '647 application (e.g., see Berger, Example D), a metal scavenger is added to achieve the highest possible safety with respect to the concentration of free Gd^{3+} ions in the formulation.

The second reason is the complexation of potentially released other metal ions from the packaging material (glass, plastics etc.) of the contrast agent formulation. Such free metal ions may also have toxic properties, similar to free Gd^{3+} ions, and their concentration is significantly reduced by immediate complexation by the metal scavenger.

10) I am a co-author of "Stability of Gadolinium-based Magnetic Resonance Imaging Agents in Human Serum at 37°." *Investigative Radiology*, **2008**, 43(12), 817-828 (previously submitted to the USPTO). In this paper, a listing of the active components in many common commercially available MRI contrast agent formulations is presented, including those that include a metal scavenging agent. In Table 1 at page 819 of this paper, commercially available gadolinium-based contrast agent formulations for MRI including excess ligand as a metal scavenger are listed (column 6, the only contrast formulations that do not include the same ligand

structure as a metal scavenger are Multihance (entry 4) and Dotarem (entry 9), both of which do not include excess ligand or other metal scavengers). Using excess ligand, either as the free ligand or its calcium complex (e.g., Calcobutrol; calcium is only weakly bound to the metal scavenger and is easily replaced by free Gd^{3+} ions) prevents accumulation of free, decomplexed Gd^{3+} ions which may be toxic in the human body. Should any Gd^{3+} become decomplexed from the gadolinium-based contrast agent, for example during shipping and storage, the free Gd^{3+} ion will be recomplexed by binding with the metal scavenger. Since the commercially available gadolinium-based contrast agents and the metal scavenger have the same ligand structure, the resulting recomplexed Gd^{3+} chelate will have the same structure as the original gadolinium-based contrast agent molecule. To this end, the '647 application describes the addition of excess DO3A-derived tetra-ligand with substoichiometric amounts of paramagnetic metal ions (i.e., less than 4 Gd^{3+} ions per ligand molecule) as a metal scavenger to reduce the concentration of free Gd^{3+} ions (see, the '647 application as filed at page 23, line 35 to page 32, line 2).

The use of metal scavengers having the same ligand structure as the gadolinium-based contrast agent is desired in the art as the active gadolinium-based chelates in the formulation *will always* have the same overall structure, regardless of whether it is the Gd chelate as originally produced or the result of decomplexation and recomplexation of the Gd^{3+} metal ion in solution with the metal scavenger. Having gadolinium-based contrast agent formulations comprising a metal scavenger with a different ligand structure could potentially have undesired impacts on the properties of the formulation since the original Gd chelate and the recomplexed Gd scavenger chelate with a different ligand structure may have different physical or chemical properties. Therefore, the use of a chemically different scavenger has never been considered or proposed in the prior art. We have demonstrated that the use of a metal scavenger with a chemical structure different to the ligand of the gadolinium-based contrast agent *does not necessarily lead to such undesired effects* (see paragraph 7) above).

As shown in the publication in Investigative Radiology, all commercially available gadolinium-based contrast agent formulations, which include a metal scavenger, use a metal scavenger structure that is identical to the ligand structure of the gadolinium-based contrast agent in the formulation. Indeed, even though Platzek established that Calcobutrol as a highly effective free Gd^{3+} scavenger for Gadobutrol formulations, manufacturers of other gadolinium-based contrast agents have not been motivated to add Calcobutrol as a metal scavenger in their formulations. Instead, these manufacturers have used metal scavengers with ligand structures that are the same as the ligand used for the gadolinium-based contrast agent, consistent with the practice in the art. I note that Gadobutrol formulations are the only commercially available gadolinium-based contrast agent formulations that include Calcobutrol. Based on the prior art and general knowledge in the art, there is no motivation to produce a gadolinium-based contrast agent formulation that includes a gadolinium-based contrast agent having a first ligand structure and a metal scavenger having a second, different ligand structure.

10) As an expert skilled in the art of the design of gadolinium-based contrast agents and pharmaceutical formulations thereof, it is my belief and opinion that one of ordinary skill in the art and based on the prior art, such as Berger and Platzek, would not be motivated to produce a gadolinium-based contrast agent formulation that includes, in part, a DO3A-derived tetra- Gd^{3+} chelate structure, as shown in Formula I in the '647 application, and a metal scavenger having a different ligand structure, such as Calcobutrol. Such a combination goes beyond the common

teaching of the art for producing formulations of gadolinium-based contrast agents and would not be obvious in view of the prior art, such as Berger and Platzek.

I hereby declare further that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with knowledge that willful statements and the like so made are punishable by fine or imprisonment, or both, under section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the above-identified application or any patent issuing thereon.

Date December 1, 2023

Thomas Frenzel
Thomas Frenzel