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WEST LAFAYETTE  
INDIANA 47906

7 August 2002

Dockets Management Branch (HFA-305) AUG 12 P2:03  
Food and Drug Administration  
5630 Fishers Lane, Room 1061  
Rockville MD 20852

Re:

*Docket No. 02D-0232*  
*Draft Guidance ICH S7B*  
*Safety Pharmacology Studies for Assessing the Potential for Delayed*  
*Ventricular Repolarization (QT Interval Prolongation) by Human*  
*Pharmaceuticals*

To Whom It May Concern:

Bioanalytical Systems Inc. is a research organization and manufacturer of instruments for chemical analysis and in vivo sampling. We are particularly interested in the development of new technologies to assist with discovery and development of new human pharmaceuticals. To this end, we feel that we have information that may be of service to the FDA when considering revisions of Draft Guidance ICH S7B.

Please allow us to provide some background information. In 1999, we introduced an automated blood sampling device which was designed to facilitate pharmacokinetics studies in rodents. This device removes programmed volumes of whole blood (from 10 to 250  $\mu$ L), at programmed time intervals, from unrestrained rats and replaces the blood volume with an equivalent volume of sterile saline. The blood is deposited into refrigerated (3°C), dark, sealed vials. The animal is free to move, sleep, groom, eat, or whatever else it chooses to do. The instrument operates 24/7 without the need for human intervention. We have demonstrated that this method of blood sampling imposes considerably less stress on the animal, as determined by comparisons in plasma or serum levels of stress hormones (e.g. epinephrine, norepinephrine, dopamine and corticosterone in rats) between rats sampled using the automated device, and the same rats sampled using conventional techniques (e.g. restraints, tail vein, jugular puncture). Blood sampling is accomplished by connecting our instrument to intravenous or intraarterial catheters in the animal. The catheters are protected from animal movement by our interactive caging system which responds to animal movement (beyond 270° of rotation) and moves the cage in the direction opposite to the animal's direction of movement, until such time as the animal stops moving. So, in addition to collecting the blood samples, the device also records animal activity, specifically rotational behavior (and more recently rearing

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behavior) as part of its standard functions. The cage is a metabolic cage, so simultaneous collection/separation of urine and feces is also accomplished.

We typically implant two catheters into each animal: one in the jugular vein (above the right atrium) and another into the femoral vein (ending up behind the left ventricle). The jugular vein catheter is used to sample blood, while the femoral vein catheter is used for intravenous dosing by bolus or continuous infusion.

Recently, we reported preliminary results on another technique which relies on the use of the automated blood sampling device described above. In essence, we converted the catheters into catheter-electrodes. They retained their function as catheters (automatically collecting blood and/or delivering drug) but we made an electrical connection to them and attached them to an electrocardiograph. Since the catheters are always filled with some type of physiological solution (saline or blood) they are also electrically-conductive. Because of their position relative to the heart, these two venous catheters therefore function as ECG leads, with recordings simulating the conventional lead III. We were sufficiently pleased by the strength of the ECG signal to explore the use of these catheter-electrodes in the rat model. We have conducted studies, in multiple rats, in which we:

- placed catheter-electrodes in the jugular and femoral veins, as described
- used a Latin Square design to dose animals with terfenadine, or ketoconazole, or vehicle, or a co-dose of terfenadine + ketoconazole
- automatically recorded 6 second electrocardiograms at intervals of every 5 minutes (note: unlike telemetry, or Holter monitors, we have virtually limitless power and can collect ECG data as long or as frequently as we need it).
- automatically collected blood samples throughout periods ranging from 4 hours to several days to establish pharmacokinetic curves for terfenadine, ketoconazole and the fexofenadine metabolite of terfenadine
- automatically recorded animal activity
- determined QT interval and calculated QTc(B) and QTc(F) and R-R

As a result of this work, we saw a very reproducible series of ECG events in rats that were dosed with the combination of terfenadine and ketoconazole. These changes did not occur when the same rat(s) were dosed with vehicle, ketoconazole alone, or terfenadine alone. The first of these was a severe depression of the R wave, followed by prolongation of the QT interval, followed by a strong suppression of the P wave, followed by return to normal state approximately 3 to 4 hours post-dose, when the terfenadine had mostly cleared from the plasma. I have attached a summary sheet with examples of these characteristic electrocardiograms.

In the draft guidance, *Section 3.1.3 In Vivo Electrophysiology Studies*, you have stated in paragraph 3 that adult rats are not suitable species. This is attributed to the differences in ionic mechanisms of repolarization in the rat. Also, in *Section 3.4.1.4 Influence of Heart Rate Change on QT Interval*, you state that "QT interval and heart rate have an inverse, non-linear relationship, which can vary among species, between animals, or even within the same animal at different heart rates".

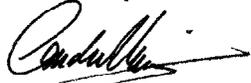
Although we are still at a relatively early stage in development of the catheter-electrode technique, we feel that it may indeed offer potential as a useful in vivo screening tool for QT interval prolongation early in drug discovery. Screening for QT interval prolongation is often difficult early in discovery due to the scarcity of compound. Rats are often candidates for first-in-animal screens, with such work focusing on early pharmacokinetics. They don't require as much drug as a rabbit, dog, pig, or other large animal models. We feel that the ability to simultaneously collect blood for PK and get a first peek at electrocardiograms from a single dose could be a useful combination. A single dose from a first-in-animal study could reveal drug-induced anomalies associated with the drug or its metabolites.

Learning more about the potential for QT interval prolongation due to a drug interaction, as in the terfenadine-ketoconazole example, would require subsequent dosing and knowledge of the routes for metabolism and excretion. Keeping in mind the scarcity of drug early in discovery, the rat could again serve as a model. In summary, we feel that the rat could be a useful early screening tool for QT interval prolongation.

We feel that the drug-induced changes in the attached electrocardiograms are sufficiently compelling to warrant further development and evaluation of the catheter-electrode technique. We propose that you reconsider the statement on use of rats in *Section 3.1.3*, instead of rejecting them completely. In particular, we would suggest that rats could be useful in early in vivo studies as a screening tool. Since ours is a novel approach, data like this was not previously available for consideration. Since we do not handle or restrain animals during these studies, the heart rate of the rats is considerably reduced, benefiting the inverse, non-linear relationship mentioned in *3.4.1.4*. Since we have no limit on battery life, we can collect a lot of electrocardiograms and this helps to capture transitional events (such as R-wave suppression, as shown). Because we can simultaneously collect blood, administer drug, and monitor behavior, we collect additional relevant information, which can be used for dose-ranging the onset of the QT prolongation effect in the same animals.

Thank you for considering information about this new technology. I would be pleased to share the data we have collected if it interests you.

Sincerely,

A handwritten signature in black ink, appearing to read "Candice B. Kissinger", with a long horizontal flourish extending to the right.

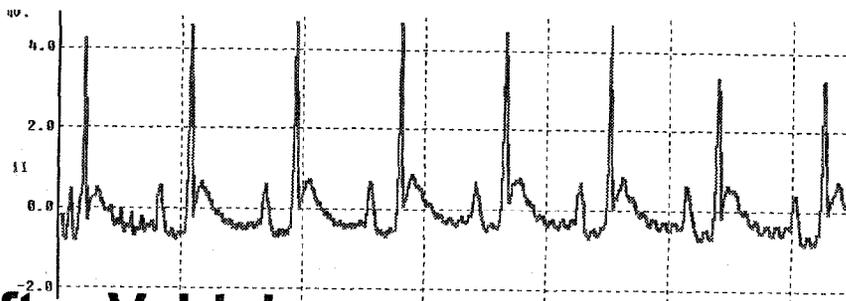
Candice B. Kissinger

Director, In Vivo Sampling

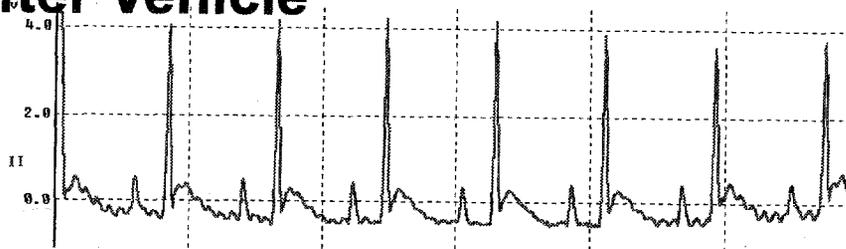
[candice@bioanalytical.com](mailto:candice@bioanalytical.com)

Same Male Rat with Catheter-Electrodes in Right Jugular Vein and Left Femoral Vein After 4 Separate Dosing Events

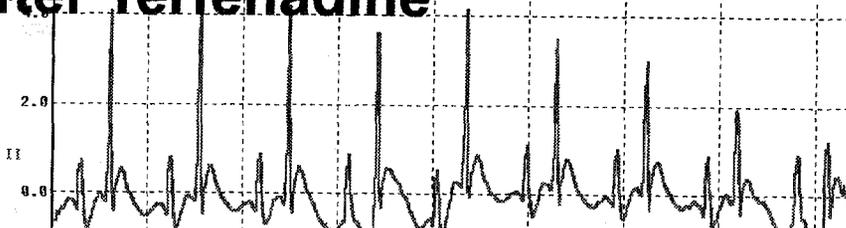
**Baseline**



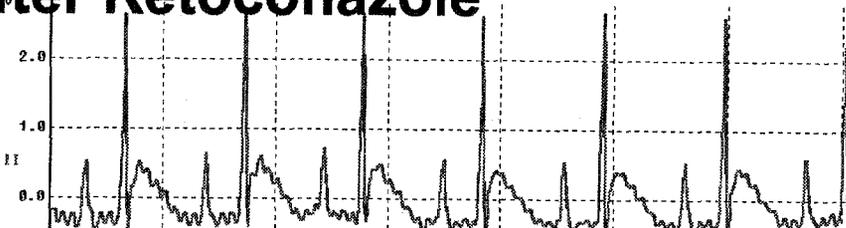
**30 min. after Vehicle**



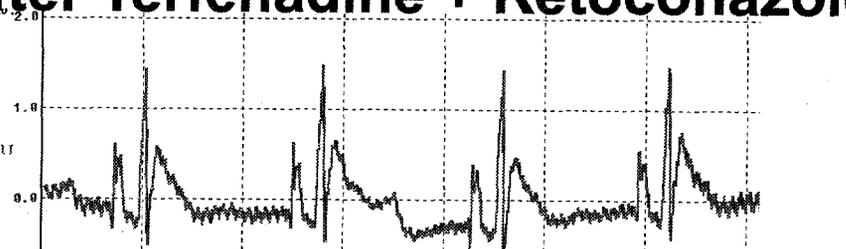
**30 min. after Terfenadine**

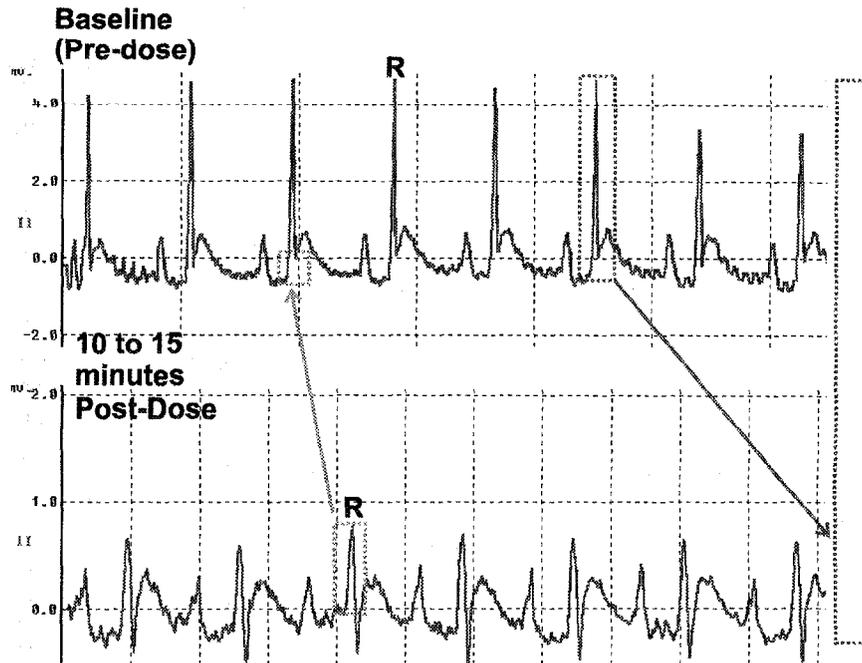


**30 min. after Ketoconazole**



**30 min. after Terfenadine + Ketoconazole**





Depression of the R wave is severe as illustrated in this comparison of baseline vs. post-dose ECG for a rat dosed with terfenadine + ketoconazole. These two electrocardiograms are shown at different scales. The dotted rectangles, connected by arrows, show how large (or small) the labeled R wave would be on the same scale as shown on the other ECG.



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