

# ECG recording methods in dog toxicology studies: what is the real benefit of external telemetry ?

Lainée P, Schofield J, Draper C, Elliott K, Barnard C and Valentin JP

AstraZeneca, Global Safety Assessment, Alderley Park, Macclesfield, United Kingdom

## Does external telemetry improve data quality ?

### Aim

ECG recordings in Toxicology studies (Tox) have historically been conducted on restrained animals, during short sessions named snapshots. Whatever the method used (i.e. standing or lying dogs, slings ...), valuable data can only be obtained if the appropriate training takes place prior to the study. In the same time, Safety Pharmacology (SP) studies have benefited from invasive telemetry recordings to acquire continuous data, enabling selection of the most appropriate time periods to assess drug effects. Using such different methods, a significant difference is observed on the amount and the quality of data, leading to question the interest and benefit of ECG recordings in Tox studies. The availability of external telemetry using jacketed animals offers an interesting alternative that has been assessed in our Tox studies for more than five years now. This review presents a comparison of historical data obtained using the three different methods.

### Methods

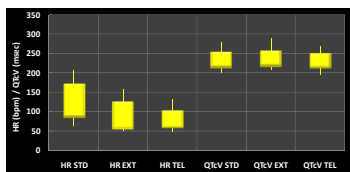
Dog data collected over the last nine years have been extracted from our historical database to compare baseline or pre-study data obtained during Tox or SP studies. To limit biases, the following criteria were used:

- For standard snapshot recordings (STD, n=314), pre-study data were obtained from trained dogs included in different study types (7 MTD/DRF and 9 pivotal 1/3 month). The acquisition system was either a paper chart recorder or a computerised system (IOX and ECG Auto, EMKA Technologies)
- For external telemetry (EXT, n=177), pre-study data were obtained from 11 MTD/DRF and 4 pivotal studies. The acquisition system was fully computerised (IOX, Study Designer and ECG Auto, EMKA Technologies)
- For standard invasive telemetry (TEL, n=111), baseline data from the pre-dose period were used, and only the first recording of the first study where the animals were included were taken in account. The acquisition system coupled the telemetry hardware (Data Sciences International)

Descriptive statistics (see table below) were obtained for heart rate (HR) and standard ECG intervals: PR, QRS, QT and QTcV (Van de Water correction). The limits of statistical significance for the different study types were calculated based on a 80% statistical power and a group of 4 (DRF or TEL studies) or 6 dogs (1/3 month studies).

## Results

Parameter	HR (bpm)			PR (msec)			QRS (msec)			QT (msec)			QTcV (msec)		
	STD	EXT	TEL	STD	EXT	TEL	STD	EXT	TEL	STD	EXT	TEL	STD	EXT	TEL
Mean	126	85	81	96	101	105	47	46	42	192	218	211	235	239	232
SD	26	20	14	13	12	11	5	8	5	16	18	12	12	12	12
SD / Mean	21%	23%	17%	13%	12%	11%	11%	18%	11%	8%	8%	6%	5%	5%	5%
Min	64	52	50	65	76	82	32	34	31	150	167	189	201	209	197
Max	205	158	130	143	147	139	67	68	57	245	291	245	276	290	268
IC5	86	56	58	76	82	87	39	38	36	168	190	194	215	219	214
IC95	171	124	101	119	122	128	56	63	51	218	247	233	253	258	249



**HR and QTcV descriptive statistics**  
Plots of minimal, maximal and confidence intervals (IC5-IC95) obtained using the three different methods. HR values are lower and less variable using external telemetry method, leaving more chance to detect HR variations. No differences are noticed for QTcV or other ECG intervals.



**Ranges of non significant HR values (statistical power analysis)**  
With narrower ranges, the use of telemetry is more likely to benefit from statistical tests. Better study conditions could explain the difference between DRF EXT and TEL, which might still benefit from staff training. The use of STD can only detect huge HR variations.

## Discussion - Conclusions

The most relevant benefit of telemetry demonstrated by this analysis is the availability of reliable and more physiological heart rates. This is both due to the long term recordings, within which the best time period around the targeted time point can be selected, and to the unrestrained conditions reducing the background stress often observed during snapshot sessions. Not much difference is observed on other parameters but HR. In such conditions, the inclusion of statistical tests might be more relevant, looking for differences from physiological baseline values, and leading to a greater chance to close the gap between statistical significance and biological relevance.

The next objective is to check whether this helps to better detect cardiac adverse effects (AEs).

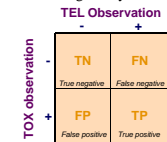
## Does it help to better detect ECG adverse events?

### Aim

The previous section demonstrated that baseline values obtained from telemetry recordings are more physiological and less variable. In addition, one would expect that measuring multiple time points facilitates the detection of variations, or at least decreases the impact of outliers when it comes to interpret data, usually when small groups are used. In this section, we aim to compare the CV findings detected by the different methods when used in similar conditions. The results obtained from the SP telemetry studies were considered as the reference. As telemetry studies routinely assess the effects after single administration, comparison were made against initial findings obtained in Tox (ideally Day 1).

### Methods

The review included 72 projects for which SP telemetry data were available. Toxicology data were obtained from 55 DRF (either 7 or 14 days) and 60 1-month studies. The ECG findings were extracted from the final reports, and adverse effects were considered as those described as relevant in the study report summary or text (with no reinterpretation of the data). Three ECG methods were separated: single snapshot (SIN: only one post-dose time point), repeated snapshots (REP: more than one post-dose time point) and external telemetry (EXT: continuous recording with multipoint extraction). The findings analysed were: increases or decreases of HR and prolongations of QTc, QRS and PR.

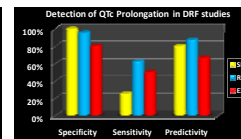
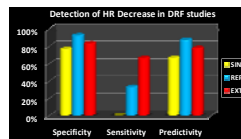
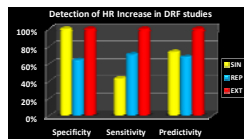


The specificity, sensitivity and overall predictivity of each ECG method vs SP telemetry was assessed as described below:

- Specificity = TN / (TN + FP) : High specificity reflects a low rate of false positives
- Sensitivity = TP / (TP + FN) : High sensitivity reflects a low rate of false negatives
- Predictivity = (TP + TN) / Total : High predictivity reflects a low rate of false responses

## Results

DRF SIN	n	TP	FP	TN	FN	Specificity	Sensitivity	Predictivity	1-Month SIN	n	TP	FP	TN	FN	Specificity	Sensitivity	Predictivity
HR Increase	15	3	0	8	4	100%	43%	73%	HR Increase	26	4	0	13	9	100%	31%	65%
HR Decrease	15	0	3	10	2	77%	0%	67%	HR Decrease	26	0	2	21	3	91%	0%	81%
QTc Prolongation	15	0	1	11	3	100%	25%	80%	QTc Prolongation	26	2	0	18	6	100%	25%	77%
QRS Prolongation	15	0	0	15	0	100%	-	100%	QRS Prolongation	26	1	0	24	1	100%	50%	96%
PR prolongation	15	0	0	14	1	100%	0%	93%	PR prolongation	26	1	0	24	1	100%	50%	96%
DRF REP	n	TP	FP	TN	FN	Specificity	Sensitivity	Predictivity	1-Month REP	n	TP	FP	TN	FN	Specificity	Sensitivity	Predictivity
HR Increase	31	12	5	9	5	64%	71%	68%	HR Increase	29	11	2	11	5	85%	69%	76%
HR Decrease	31	1	2	26	2	93%	33%	87%	HR Decrease	29	1	0	27	1	100%	50%	97%
QTc Prolongation	31	5	1	22	3	96%	63%	87%	QTc Prolongation	29	5	2	20	2	91%	71%	86%
QRS Prolongation	31	1	3	27	0	90%	100%	90%	QRS Prolongation	29	0	3	26	0	90%	-	90%
PR prolongation	31	3	0	27	1	100%	75%	97%	PR prolongation	29	2	0	24	3	100%	40%	90%
DRF EXT	n	TP	FP	TN	FN	Specificity	Sensitivity	Predictivity	1-Month EXT	n	TP	FP	TN	FN	Specificity	Sensitivity	Predictivity
HR Increase	9	3	0	6	0	100%	100%	100%	HR Increase	5	0	0	4	1	100%	0%	80%
HR Decrease	9	2	1	5	1	83%	67%	78%	HR Decrease	5	0	1	3	1	75%	0%	60%
QTc Prolongation	9	2	1	4	2	80%	50%	67%	QTc Prolongation	5	0	1	4	0	80%	-	80%
QRS Prolongation	9	1	1	7	0	88%	100%	89%	QRS Prolongation	5	0	0	5	0	100%	-	100%
PR prolongation	9	0	1	7	1	88%	0%	78%	PR prolongation	5	0	0	5	0	100%	-	100%



**Impact of the different methods when used in DRF studies**

With larger ranges of baseline HR values, relevant increases or decreases of HR are not often observed using single snapshots, leading to poor sensitivity. On the other hand, when such an effect is observed, it is likely to be a reliable, hence the good specificity. In all cases, repeated measurements improve sensitivity, with more obvious difference observed EXT.

## Discussion - Conclusions

The first observation is that overall predictivity is good, in spite of very limited number of animals in the DRF studies (n=2 to 4), with predictive values often greater than 80%.

The use of repeated measurements improves sensitivity, whatever the method used. This might correlate with a stronger confidence when moderate effects are observed repeatedly as opposed to only once where an outlier cannot be ruled out. The use of EXT adds to this confidence with narrower HR baseline ranges, and more chance to reach the threshold for a relevant effect to be considered.

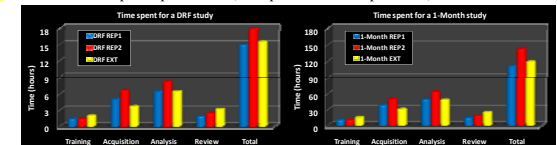
PR/QRS prolongations were rarely observed from SP telemetry studies and no clear conclusion can be drawn.

From this analysis, there is a clear benefit to include repeated measurements in Toxicology studies, therefore reaching an appropriate level of confidence in the effects detected. The best method to adopt is then dependent on strategic choices and investment decisions. Practical aspects such as cost or time investment are key components to the decision.

## Why and when should we use it in Tox studies?

### Comparison of time requirements

Assuming that repeated measurements are required, the standard times needed to generate data using snapshots (REP1: 2 post-dose recordings, REP2: 3 post-dose recordings) or external telemetry (EXT) were compared. All components of data generation (training, acquisition, analysis and quality control) were measured and reported as amount of hours spent using a standard designs (1 early and 1 late session during study, 1 during recovery) in DRF (4 dogs) or 1-month studies (30 dogs).

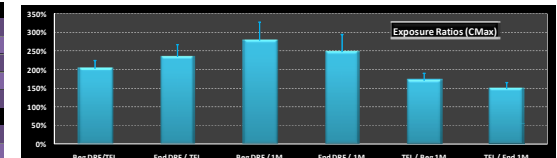


If repeated recordings include more than 2 time points post-dose, the use of external telemetry becomes beneficial. The slightly longer time required for reviewing / checking the data is compensated by the faster acquisition phase, as animal handling is limited to jacket installation and removal.

### Comparison of exposures between study types

Safety margins used during risk assessment are routinely calculated using plasma exposures. When evaluation of cardiovascular adverse effects (AEs) are purely based on SP telemetry studies, exposure are limited to those obtained after single administrations. Increasing the range of exposures will either facilitate detection of the first AEs or increase the safety margin if the compound is free of AEs.

Maximal exposures (Cmax) obtained at the beginning and the end of the repeated Tox studies were compared them to those from SP studies.

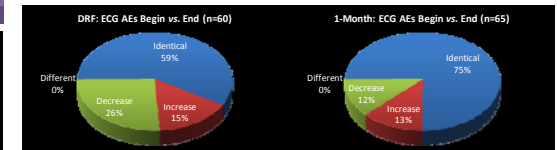


Exposures reached in the DRF studies is about twice higher than in the telemetry studies, for which does not exceeding the MTD are selected to limit clinical signs and facilitate re-use of the animals. SP telemetry-exposures are however higher than those obtained in the pivotal 1-month studies. The exposure ranking is therefore: DRF > TEL > 1-Month

### Modification of ECG findings after repeated administrations

Finally, a comparison was made between findings generated during the early phase of the Tox studies (i.e. first days of dosing) and those obtained at the end of the studies (i.e. either after 7, 14 or 28 days of repeated dosing).

Four categories were defined whether the effects were: identical, increased (magnitude of the AE increased or additional ones described), decreased (magnitude of the AE decreased or less AEs observed) or different (AEs observed initially replaced by a different one). Results are presented as percentage of the number of studies.



The effects observed at the study end after repeated administrations are different in about 40% and 25% of the cases in the DRF and 1-month studies respectively. When it comes to correlate these findings with either clinical or pathological ones along the study, single dose investigations could be insufficient and even misleading.

## Conclusions

This review demonstrates a) that ECG recordings performed in Toxicology studies are valuable, b) that inclusion of multiple time points improves the sensitivity of the ECG analysis and c) that external telemetry provides the most reliable values, similar to those obtained in invasive telemetry studies.

External telemetry should therefore be considered as the gold standard in Tox as invasive telemetry in SP designs, and be used as such as early as possible during development, for example in DRF studies. The method is not more time consuming than repeated snapshots, and the same software selected for SP studies can be used, therefore enabling sharing expertise, optimising resources and ultimately reducing cost.

External telemetry is especially valuable when Safety Pharmacology ECG end points must be obtained from Toxicology studies. This might facilitate application of the recent ICH S9 guideline, enabling some oncology compounds to be tested as part of the pivotal Toxicology studies. Similarly, external telemetry is very beneficial for biologics development, according to ICH S6 recommendations: with non-human primates as the preferred species, ECG recordings from restrained animals are in deed poorly informative and external telemetry should be considered to provide the relevant regulatory data.