



# PHOSPHORUS KIT

(Molybdate U.V. Method)

(For veterinary invitro diagnostic use only)

### INTENDED USE

QUADRAPED™ Phosphorus kit is used for the determination of Inorganic Phosphorus in serum or plasma.

### SUMMARY

Phosphorus is mainly combined with calcium and is found in the bones. Approximately 15% exist as inorganic phosphorus or phosphate esters. It is involved in the carbohydrate metabolism and is a component of many other substances. Increased levels are found in hypoparathyroidism, renal failure, bone metastasis and liver diseases. Decreased levels are found in hyperparathyroidism, rickets and Vitamin D deficiency.

### PRINCIPLE

Phosphate ions in an acidic medium react with ammonium molybdate to form a phosphomolybdate complex. This complex has an absorbance in the ultraviolet range and is measured at 340 nm. Intensity of the complex formed is directly proportional to the amount of inorganic phosphorus present in the sample.



### EXPECTED VALUES

Species	Phosphorus (mg/dl)
Dog	2.9 - 5.3
Cat	3.0 - 6.1
Cow	5.6 - 8.0
Horse	1.5 - 4.7
Pig	5.3 - 9.6
Sheep	5.0 - 7.3
Goat	4.2 - 9.1
Rabbit	4.0 - 6.5
Buffalo	4.0 - 7.5

It is recommended that each laboratory establish its own range as reference ranges may vary between laboratories.

### PRESENTATION

REF	1126170025
Pack Size	25 ml
L1 Acid Reagent	20 ml
L2 Molybdate Reagent	5 ml
S Phosphorus Standard (5 mg/dl)	5 ml

### COMPOSITION

Sulphuric Acid 700mM; Ammonium Molybdate 0.4mM; Detergent.

### STORAGE / STABILITY

Contents are stable at 23-29°C till the expiry mentioned on the labels.

**Working reagent:** Pour the contents of 1 bottle of L2 (Molybdate Reagent) into 1 bottle of L1 (Acid Reagent). This working reagent is stable for at least 6 months when stored at 2-8°C. Upon storage the working reagent may develop a slight blue colour however this does not affect the performance of the reagent. Alternatively for flexibility as much of working reagent may be made as and when desired by mixing together 4 parts of L1 (Acid Reagent) and 1 part of L2

(Molybdate Reagent). Alternatively 0.8 ml of L1 and 0.2 ml of L2 may also be used instead of 1 ml of the working reagent directly during the assay.

### SAMPLE MATERIAL

Serum, EDTA / Heparin / Citrated plasma. Inorganic phosphorus is reported to be stable in serum/ plasma for 7 days at 2-8°C.

### SAMPLE WASTE AND DISPOSAL

Do not reuse the reagent containers, bottles, caps or plugs due to the risks of contamination and the potential to compromise reagent performance.

Appropriate biosafety practices should be used for materials that contain or are suspected of containing infectious agents.

Handle specimens, solid and liquid waste and test components in accordance with local regulations and NCCLS guidelines M29, or other published biohazard safety guidelines.

### MATERIALS REQUIRED BUT NOT PROVIDED

Photometer analyzer with standard thermostatic cuvette holder, micropipette and appropriate laboratory equipment.

### PROCEDURE

Wavelength / filter : 340 nm (Hg 365 nm)  
Temperature : R.T.  
Light path : 1 cm

Pipette into clean dry test tubes labelled as Blank (B), Standard (S) and Test (T):

Addition Sequence	B (ml)	S (ml)	T (ml)
Working Reagent	1.0	1.0	1.0
Distilled Water	0.01	-	-
Phosphorus Standard (S)	-	0.01	-
Sample	-	-	0.01

Mix well and incubate at R.T. for 5 mins. Measure the absorbance of the Standard (Abs. S), and Test Sample (Abs. T) against the Blank, within 60 mins.

### CALCULATIONS

$$\text{Phosphorus in mg/dl} = \frac{\text{Abs.T}}{\text{Abs.S}} \times 5$$

### QUALITY CONTROL

The following process is recommended for QC during the assay of Phosphorus. \*Define and establish acceptable range for your laboratory.

- Two levels of control (Normal and Abnormal) are to be run on a daily basis.
- If QC results fall outside acceptance criteria, recalibration may be necessary.
- Review QC results and run acceptance criteria following a change of reagent lot.

### SPECIFIC PERFORMANCE CHARACTERISTICS

LOD: 0.1 mg/dl

LOQ: 0.62 mg/dl

LOB: 0.1 mg/dl

Lower Limit: 0.1 mg/dl

Higher Limit: 20 mg/dl

If values exceed this limit, dilute the sample with distilled water and repeat the assay. Calculate the value using the proper dilution factor.

### Interferences:

Sample when spiked with interferent such as upto 10 mg/dl Bilirubin, 1000 mg/dl intralipid and 200 mg/dl haemoglobin does not affect the ability of the kit to determine Phosphorus concentration.

### Precision:

#### Within run

Within run	n	Mean	SD	% CV
Sample 1	10	5.49	0.06	1.15
Sample 2	10	7.53	0.07	0.88
Sample 3	10	16.46	0.49	2.95

#### Between run

Between run	n	Mean	SD	% CV
Sample 1	10	5.46	0.06	1.06
Sample 2	10	7.56	0.08	1.11
Sample 3	10	16.46	0.48	2.92

### Method comparison:

Comparative studies were done to compare our reagent with another commercial Phosphorus Assay. No significant differences were observed. Details of the comparative studies are available on request.

### NOTE

In vitro diagnostic reagent for laboratory and professional use only Not for medicinal use. The reagent contain sodium azide 0.1% as preservative. Avoid contact with skin and mucosa. On disposal flush with large quantities of water. Only clean and dry glassware must be used. Hemolysis interferes with the test.

Use clean glassware washed with N/10 HCl as detergents may contain phosphate ions. Do not use turbid, deteriorated or leaking reagents.

### REFERENCES

- Fiske, C.H., Subbarow, Y. (1925) J. Biol. Chem. 66 : 375.
- Goodwin, J.F., (1970) Clin. Chem. 16(19) : 776.
- Henry J. R. (1964) P. 415 in J. R. Henry Clinical Chemistry.
- Duncan and Prasse's Veterinary Laboratory Medicine: Clinical Pathology, Kenneth S. Latimer, ISBN Jane Wardrop, 6th Edition - 2010
- Clinical Biochemistry of Domestic Animals, Sixth Edition, 2008 by Kaneko J.J., Harvey J.W. & Bruss M.L.
- Data on file: Coral Clinical Systems.

### System Parameters

Reaction	: U.V. End Point	Interval	: —
Wavelength	: 340 nm	Sample Vol.	: 0.01 ml
Zero Setting	: Reagent Blank	Reagent Vol.	: 1.00 ml
Incub. Temp.	: R. T.	Standard	: 5 mg/dl
Incub. Time	: 5 min.	Factor	: —
Delay Time	: —	React. Slope	: Increasing
Read Time	: —	Linearity	: 20 mg/dl
No. of read.	: —	Units	: mg/dl

### SYMBOL KEYS

Store at 23-29°C	Manufacturer	<b>IVD</b> In vitro Diagnostic Medical Device	<b>L1</b> Acid Reagent	<b>Molybdate U.V.</b> Molybdate U.V. Method
Use by (Last day of stated month)	Consult Instructions for use	<b>LOT</b> Batch Number	<b>L2</b> Molybdate Reagent	<b>S</b> Phosphorus Standard (5 mg/dl)
Date of Manufacture	<b>REF</b> Catalogue Number			This way up



Manufactured by:

## Coral Clinical Systems

A Division of Tulip Diagnostics (P) Ltd.

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