

# Application News

Spectrophotometric Analysis

No.A429

## Leach Test for Phenol in Rubber Nipples

As food moves through the process starting from the raw material stage to the point of ingestion, it comes into contact with a wide variety of items, including implements, containers and packaging materials. These points of physical contact can include equipment used for manufacture and processing, containers and packaging materials used for storage and transport, and cooking and eating utensils used in restaurants and in the home.

Because these implements, containers and packaging materials are made of various types of materials, such as rubber, glass, and metals, there is always the possibility that their constituent substances, as well as impurities, can be taken into the body via the ingested food. Therefore, it is necessary to secure the safety of

implements, containers, and packaging materials as provided for in the Japan's Food Sanitation Act, "Standards and Criteria for Food and Food Additives, etc., Chapter 3: Apparatus and Containers and Packaging."

One example of a rubber implement is the rubber nursing nipple, which is inserted directly in an infant's mouth. Since it is possible that toxic phenol can leach out of the rubber nipple when it is inserted into an infant's mouth, a leach quantity standard has been established in the Food Sanitation Act, and testing is specified to be conducted using an UV-VIS spectrophotometer. Here we introduce the phenol leach test for quantitation of phenol leached from a rubber nipple according to the Food Sanitation Act.

### ■ Standard for Implements, Containers and Packaging for Foods

Table 1 lists the target analytes and analytical instruments to be used for measurement with respect to the various types of implements and packaging materials. Both general standards and specific standards are specified for rubber and plastic implements, or packaging and containers. Phenol is indicated as an analyte for metal cans, for rubber in the general standard as well as the nursing nipple specific standard, and for phenol plastic, melamine plastic and urea plastic specific standard. In all of these cases, an UV-VIS spectrophotometer is specified for conducting measurement.

### ■ Preparation of Reagents

The various reagents are prepared as described below.

- Phenol standard solution  
Dissolve 1.0 g of phenol in water, and adjust the volume to 100 mL. Take 1 mL of this solution, and add water to bring the volume to 100 mL. Take 1 mL of this solution, and add water to bring the volume to 20 mL. (1 mL of this solution contains 5 µg of phenol.)
- Borate buffer solution  
1st solution: Dissolve 4.0 g of sodium hydroxide in water, and adjust the volume to 100 mL.  
2nd solution: Dissolve 6.2 g of boric acid in water, and adjust the volume to 100 mL.  
Mix equal volumes of the 1st solution and the 2nd solution.
- 4-aminoantipyrine reagent  
Dissolve 1.36 g of 4-aminoantipyrine in water, and adjust the volume to 1000 mL.
- Potassium hexacyanoferrate (III)  
Dissolve 8.6 g of potassium hexacyanoferrate (III) in water, and adjust the volume to 1000 mL using 1.8 mL of aqueous ammonia (28 to 30 % concentration) and water.

Table 1 Target Analytes and Analytical Instruments

Implement / Packaging and Container	General /Specific Standard	Target Analyte Substances	Analytical Instrument <sup>Note 1)</sup>
Glass, earthenware, and enamel	-	Cd, Pb	AA/ICP
Metal cans	-	Cd, Pb	AA/ICP
		Phenol	UV-VIS
		Epichlorohydrin	GC
		Vinyl chloride	GC
Rubber	General standard	Cd, Pb, Zn	AA/ICP
		Phenol	UV-VIS
	Rubber nipples	Cd, Pb, Zn	AA/ICP
		Phenol	UV-VIS
Plastics	General standard	Cd, Pb	AA/ICP
	Phenol plastic Melamine plastic Urea plastic	Phenol	UV-VIS
	Polyvinyl chloride	Dibutyltin compounds	GC/MS
		Cresol phosphate esters	HPLC
		Vinyl chloride	GC
	Polystyrene	Volatile substances (5 types)	GC, GC/MS
	Polyvinylidene chloride	Vinylidene chloride	GC
		Ba	AA/ICP
	Polyethylene terephthalate	Sb, Ge	AA/ICP
	Polymethylmethacrylate	Methyl methacrylate	GC
	Nylon	Caprolactam	GC
	Polycarbonate	Amines	GC
		Bisphenol A, diphenyl carbonate	HPLC
Poly lactate	Total lactate	HPLC	

Note 1: AA stands for Atomic Absorption spectrometer, ICP for inductively coupled plasma emission spectrometer, UV-VIS for ultraviolet-visible spectrophotometer, GC for gas chromatograph, GC/MS for gas chromatograph - mass spectrometer, and HPLC for liquid chromatograph.

### ■ Pretreatment and Analytical Conditions

Using a ratio of 1 g of sample to 20 mL of water, immerse the sample in water. Cover the receptacle with a glass plate and set aside for 24 hours, maintaining a temperature of 40 °C. Use this leach solution as the test solution.

Add 3 mL of the borate buffer solution to 20 mL of the test solution, and after shaking it well to thoroughly mix the solutions, add 5 mL of the 4-aminoantipyrine solution and 2.5 mL of the potassium hexacyanoferrate (III) solution. Add water to bring the volume to 100 mL, and after shaking well to mix the solutions, set the solution aside for 10 minutes at ambient temperature. Separately, take 20 mL (5 µg/mL) of the phenol standard solution, and perform the same procedure as that for the test solution.

Here we used the UV-1800 UV-VIS spectrophotometer, and conducted measurement using the analytical conditions shown in Table 2.

**Table 2 Analytical Conditions**

Photometric Value	Absorbance
Slit Width	1.0 nm
Wavelength Range	300 to 700 nm
Scan Speed	Medium
Sampling Pitch	1 nm

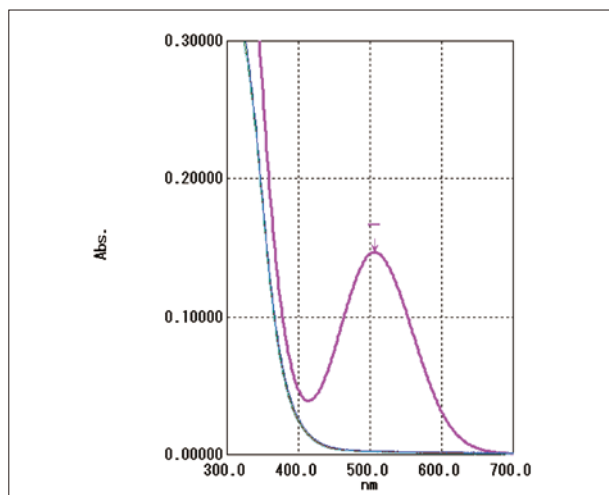
### ■ Results

After preparing test solutions as described above for four types of commercially available rubber nipples, in addition to the standard solution, measurement was conducted using the UV-VIS spectrophotometer. The absorbance spectra obtained for each test solution and the standard solution are shown in Fig. 1, and the phenol contents indicated by the absorbance values at 510 nm are shown in Table 3. Extremely low absorbance values were obtained with all of the rubber nipples.

With absorbance values lower than that of the specified value for leached phenol, the test solutions here showed almost no leaching of phenol, thereby confirming that the specified standard was satisfied.

This Application News introduced the phenol leach test for rubber nipples, however, it must be noted that some of the phenol leach conditions are different for types of rubber other than that used in nipples, as well as for metal cans and plastics.

In this way, an UV-VIS spectrophotometer can be used for conducting the leach test of phenol from implements, containers, and packaging material used for food.



**Fig. 1 Spectra of Standard Solution and Sample Solutions**

**Table 1 Analytical Conditions**

Sample Name	Absorbance (510 nm)
Company A: isoprene rubber	0.003
Company A: silicone rubber	0.002
Company B: isoprene rubber	0.002
Company B: silicone rubber	0.002
Standard solution	0.146

#### Reference:

Author: Yoko Kawamura, "Standards and Criteria for Apparatuses, Containers and Packaging, March 2006 Revision" Chuohoki Publishing



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