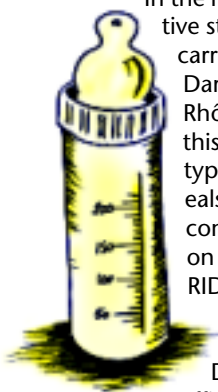


Deoxynivalenol in Baby Food

Since infants and small children need special protection, last year Germany suggested to the EU Commission that limit values for baby food and food for small children should be established for important mycotoxins such as ochratoxin A, deoxynivalenol (DON), fumonisin B₁ and B₂, zearalenon and patulin. For DON 100 µg/kg (ppb) in cereals, cereal products and noodles used for baby food was notified.

In the recent weeks a comparative study on this subject was carried out at R-Biopharm in Darmstadt and R-Biopharm Rhône Ltd. in Glasgow. In this study eight different types of baby food and cereals were tested. The DON contents were determined on the one hand using the RIDASCREEN® DON test, with and without purification using DONprep (immunoaffinity columns), and also using a combination of DONprep + HPLC. In these tests a DON content significantly below 100 ppb was found in all the samples. Seven of the eight samples had a DON content of < 37 µg/kg (ppb) [detection limit of the test: standard 2 x dilution factor], analyzed with the RIDASCREEN® DON-ELISA without DONprep. A content of



45 ppb was determined in one sample with the ELISA and confirmed with 38 ppb (DONprep + ELISA). The DONprep + HPLC analysis gave a content of 33 ppb. On the whole the ELISA results were slightly higher than the HPLC results. The recovery rate of the samples spiked with 100 ppb was between 97 and 135 % with the ELISA. The cereal samples (wheat, barley, oats, rye and sorghum) which are often used in baby food were also analyzed using the RIDASCREEN® DON test. All the samples, except the oats sample, were significantly below the detection limit. The oats sample had a low DON content of 31 ppb. With a combination of DONprep + ELISA 22 ppb were measured. In the samples spiked with 100 ppb the recovery rate was between 82 and 109 %, determined with ELISA without immunoaffinity column.

The comparative study showed that the RIDASCREEN® DON test is very suitable as a screening test for baby food while the combination of DONprep immunoaffinity columns + HPLC can be used as a confirmatory method.

The results of this study were presented in a suitable poster at the 2nd World Mycotoxin Forum on 17th and 18th February in Noordwijk aan Zee in Holland. A copy of this poster can be obtained from R-Biopharm on request.


FAPAS® Study - Aflatoxin M₁ (Series 4, Round 50, January 2003)

An established principle for assessment of the quality of analytical tests is the participation in so-called proficiency tests within the framework of the FAPAS® studies (Food Analysis Performance Assessment Scheme). These studies are organized by the Proficiency Testing Group of the Central Science Laboratory, London, UK. This laboratory is under the control of the British Ministry of Agriculture, Fishery and Food, and is therefore closely connected with the

European Commission. Every year approx. 120 studies of this kind are carried out with different analytical parameters and sample materials.



At the end of last year the detection of aflatoxin M₁ in milk was checked within the framework of a FAPAS® study. A total of 65 laboratories took part in this



ring study, each using the usual laboratory methods.

Eight laboratories used the RIDASCREEN® Aflatoxin M1 test in this study. The specified value of 0.059 µg/kg (ppb) was easily achieved on average. The mean value of the results was 0.063 ppb with a standard deviation of 0.004, a

coefficient of variation of 6.2 % and a recovery rate of 107.3 %.


Most of the laboratories used products from R-Biopharm Rhône Ltd. (Aflaprep M + HPLC). The results determined using this method corresponded almost exactly to the specified value at approx. 0.056 ppb (recovery rate: approx. 96 %).

About Our Products

RIDASCREEN® Chloramphenicol


Chloramphenicol (CAP) is a wide-range antibiotic which has been frequently used in animal production owing to its excellent antibacterial and pharmacokinetic properties. Since 1994 its use as an animal drug has been prohibited and, according to Council Directive 2377/90 (EEC), Appendix IV, zero tolerance applies in the European Union. The active substance, chloramphenicol, is suspected of causing aplastic anaemia in humans when taken therapeutically in the mg and g range, through damage of the bone marrow. However, the threshold value for triggering aplastic anaemia in humans is not known. From genotoxicity tests and toxicity studies used for assessing chloramphenicol it was concluded that no ADI (Acceptable Daily Intake Value) could be established and the drug was included in Appendix IV, according to Article 5 of Directive 2377/90 (EEC).

In the last two years monitoring of food for chloramphenicol has been considerably intensified in accordance with the Council decision of 19th September 2001 (2001/699/EC), following repeated detection of CAP residues in prawns from South-East Asia. Both aqua culture prawns and free living prawns were affected. Apart from the prawns mentioned, increased chloramphenicol contents have also been found in food such as shrimps, fish, meat and honey imported from China. CAP residues have also been discovered in products from Thailand, Vietnam and Indonesia.



Owing to an increased demand in recent months for CAP analyses in various food samples, R-Biopharm AG has developed or optimized several supplementary sample preparation methods. In addition to CAP detection in milk [German Food Law (LMBG), § 35 - Methods], meat and eggs, as described in the RIDASCREEN® product information leaflet on chloramphenicol, we also offer optimized methods for milk, milk powder and a method for shrimps, meat and fish meal. These methods have been considerably improved with regard to their detection limit so that very sensitive methods for CAP analysis are now available. Furthermore, we supply sample preparation methods for CAP in honey, mixed animal food, serum/plasma and CAP-glucuronide in urine. These supplementary sample preparation methods can be obtained from your local distributor on request.

Since no binding analytical methods for the determination of CAP exist at present, various methods (HPLC, GC-MS/NCI, GC-ECD, LC-MS/MS and ELISA) are currently used. Within the framework of a permanent veterinary committee for the harmonisation of CAP analysis a minimum performance limit of 0.3 µg/kg has been fixed by representatives of the EU member states – although this has not been legally established. This value is taken as the decisive limit for product complaints in most EU countries, and frequently in the individual federal states of Germany. However, this procedure is not uniform.



In November 2002 a comparative laboratory study on chloramphenicol was initiated by the German Seafood Association [Deutscher Seafood Verband e.V. (DSFV)], and this was assessed by the Institute for Fishery Quality [Institut für Fischereiqualität (IFF(Q))] of the BFA for Fishery. The objective of this study was to obtain an overall view of the reliability of methods established in laboratories for the determination and quantification



of CAP residues in prawn samples. Comparison of the ELISA and GC-MS methods showed that the ELISA method gave slightly higher values on average than the GC-MS/NCI method. Basically, it was noted that the ELISA method is a screening method used for positive detection of CAP, and its most important criterion is to give as few false negative results as possible (less than 5 %). Summing up, the conclusion from this study was that a CAP content of 0.1 µg/kg (ppb), with limitations, and a CAP content of 0.3 µg/kg is currently a generally acceptable maximum value, causing few analytical problems.

In November 2002 an FAPAS® round was also held (series 02, round 41). Sample material from prawns with a content of approx. 1 µg/kg (ppb) were used. The result showed that the ELISA tests and

the chromatographic methods were practically equivalent with regard to their accuracy and reproducibility for this CAP content.

A further comparative study which was carried out by NRC (Nestlé Research Center) in Lausanne, was concerned with CAP in honey and compared the RIDASCREEN® Chloramphenicol test with a HPLC-MS/MS method. Sixteen samples with various CAP contents were tested and very good agreement between the two methods was found.



RIDASCREEN® Chloramphenicol - Test Procedure

In the past, several customers have reported positive results with the RIDASCREEN® Chloramphenicol test which could not be confirmed. Even the buffer alone already showed contamination with chloramphenicol. We investigated this problem and it was found that buffer 1, included in the test kit (for dilution of standard solution and solutions of conjugate, antibody and sample), was contaminated with chloramphenicol.

This impurity could only have been introduced by incorrect pipetting (i.e. not using new pipette tips when diluting the standard).

We would therefore like to emphasize in particular that work must be carried out carefully, and new pipette tips always used in order to avoid contamination of the reagents.

RIDA® ATP Detection System - New ATP System for Hygiene Control

Mass production of food creates big challenges for industry today. Any undesirable contamination leads to a reduction in quality of the product, time losses in production and, particularly important, delays or hindrance in release of the product. The most unpleasant consequence for the manufacturer is recall of the product.

On account of these factors it is clear that a monitoring and control system is necessary in order to achieve the required quality standards for the products. Of prime importance here is a risk analysis of the critical control points (HACCP).

Any kind of food residue, whether visible or invisible, promotes growth of microorganisms and therefore risk of contam-

ination of the products. Systems such as the ATP bioluminescence are very popular today and justifiably have a reputation of being suitable monitoring systems. Acceptance of hygiene monitoring of this kind depends mainly on simplicity of use, costs, user-friendliness and specifications. One of the most critical specifications is sensitivity.

In the RIDA® ATP Detection System simple test procedures, sensitivity and low cost have been successfully combined.

The principle of this method is the quantitative detection of intra- and extracellular adenosintriphosphates (ATP) by means of the Luciferin-Luciferase reaction. Depending on the intensity of cleansing and sterilization different ATP

concentrations of microorganisms and incompletely removed product residues are left behind on the surfaces. These can be quickly and simply detected and documented by a simple test using the RIDA® ATP Detection System. Immediately after cleansing and sterilization, controls of the measures can be carried out with ATP bioluminescence. The results can be documented and according to the results suitable measures can be initiated.

The RIDA® ATP Detection System consists of two modules - the evaluation

system and the materials used. The evaluation system consists of two luminometers - the „Berthold Vega“ and the „Berthold Junior“. Materials for use with the system include the extraction reagent in a spraying bottle, ATP-free sterile swabs, reagent tubes coated with Luciferin and Luciferase (patented reagent film) and ATP standards (each containing 1 ng ATP) for checking the apparatus.

If you are interested in the RIDA® ATP Detection System ask your local distributor for suitable product information.

About Ourselves

Removals

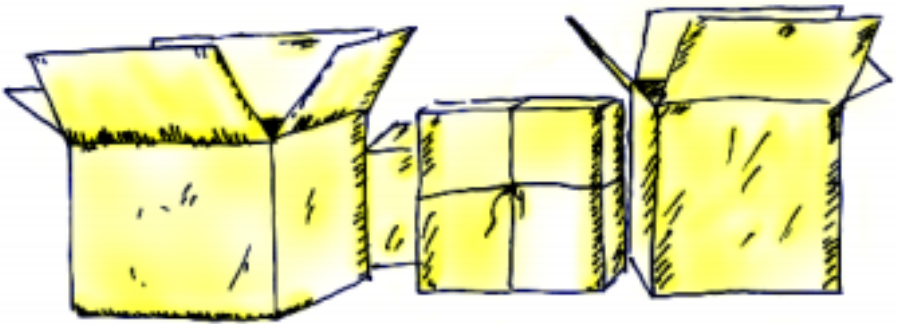
R-Biopharm moved into a new building at the beginning of March. As far as you are concerned only the name of the road has changed - otherwise everything else remains unchanged. Telephone numbers have not been changed either and you can still reach your particular contact under the same telephone number.

Our new address is as follows:

R-Biopharm AG
Landwehrstr. 54
64293 Darmstadt

(061 51) 81 02-0

We apologize in advance for any inconvenience which you may have experienced in trying to contact us at the beginning of March, or if in any other way things didn't work out as smoothly as normal.



The next RIDA News will be published during the II. quarter 2003

RIDA News is edited by
R-Biopharm AG, Dolivostraße 10, 64293 Darmstadt, Germany
phone: +49 61 51/ 81 02 - 0 (-25), fax: +49 61 51/ 81 02 - 40