

Doctoral Dissertation Abstract...

STUDIES CONDUCTED FOR THE QUANTITY DETERMINATION OF SYNTHETIC DYES ADDED INTO SOME FOOD STUFFS

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This study has been planned and carried out with a view to discover whether or not those synthetic dyes which are not allowed to be added into the jams, have already been added into the jams and also the quantities of synthetic dyes existing in the puddings, candies and granulated powder drinks which are permitted to be added with such dyes, are also compatible with the quantities specified by the Food Additives Regulation in Turkey.

All samples used during this study, have been obtained from sources of Ankara market and totally 263 samples have been analysed.

The extraction process of all samples, has been performed through the wool coloring method. The extracted dyes have been subjected to the qualitative analysis through the TLC (Thin Layer Chromatography) Method. At the end of qualitative analysis performed on the jams and puddings, it has not been possible to handle the quantitative determinations since no synthetic dye has been found.

The samples of candy and granulated powder drinks, in the contents of synthetic dyes, have determined the use of a single C₁₈ Sep-pak Cartridge and also the spectrophotometric methods.

Among these synthetic dyes, the average level of Ponceau 4R has been found as 117.45±19.37 mg/kg, in the candies, 294.79±26.21 mg/kg for the granulated powder drinks. The values were not suitable by the Food Additives Regulation.

The average level of tartrazine has been determined as 147.77±20.51 mg/kg and 201.19±37.16 mg/kg respectively for candies and granulated powder drinks. The level of tartrazine were in maximum values. The average levels of Sunset Yellow F.C.F. have been found as 174.58±31.54 mg/kg and 293.31±24.19 mg/kg respectively granulated powder drinks. The values also had been found over dose.

The average level of azorubine, has been determined as 181.22±20.22 mg/kg for candies and found compatible with those specified by the Food Additives Regulations.

The average value of mixed dyes existing in the granulated powder drinks, has been determined as 241.44 mg/kg. This value was over the value specified by the Food Additives Regulation.

A STUDY OF *LEGIONELLA PNEUMOPHILA* SEROGRUB1 ANTIGEN IN DETECTION THE URINE OF LOWER RESPIRATORY TRACT INFECTED PATIENTS USING ENZYME IMMUNO ASSAY METHOD

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The aim of this study is to describe whether the Enzyme Immuno Assay (EIA) method taking in to consideration its sensitivity, specificity and utility. It can become and alternate to the method of culture in the prescription and definition of lower respiratory tract infection due to *L. pneumophila* SG1.

For this purpose, it has been studied in lower respiratory tract infected patients' sputums, bronchoalveolar lavage (BAL) liquids and urines. *L. pneumophila* SG1 isolation with selective BCYE agar culture was made on 59 men and 34 women's sputums and BAL liquids. Again, with the EIA Urine Antigen Kit, *L. pneumophila* SG1 urine antigen has been studied in this patients' urine.

It was found that *L. pneumophila* SG1 has been grown in 2 (3.40%) of 59 men and in 1 (2.95%) of 34 women. In 2 (100%) of the urine 2 men, in whom *L. pneumophila* SG1 was grown in culture antigen was found with EIA method, while in 2 (3.50%) of the other 57 men's urine that is not growing *L. pneumophila* SG1 in culture antigen was found. In 1 (100%) of 1 woman that is growing *L. pneumophila* SG1 in culture and in 1 (3.03%) of the other 33 women not growing *L. pneumophila* SG1, antigen was found with EIA method.

In patients with lower respiratory tract infection men and women correlation rate between culture and EIA of results was found 73% and 70%.

In lower respiratory tract infection patients from *L. pneumophila* SG1, 20.43% *S. pneumoniae*, 5.37% *S. aureus*, 4.30% *H. influenzae*, 5.37% *E. coli*, 2.15 % *K. pneumoniae*, 2.15 % *P. aeruginosa*, 2.15 % *B. catarrhalis* and 1.07 % fungi have been found.

In EIA test when the culture method is taken basis, have 100% sensitivity and 96% specificity. In case, a standard EIA method is prepared, can become an alternative to the cultural identification early diagnosis of *L. pneumophila* SG1 infections and it is method that is also reliable, easily applicable and giving fast results. In this way, we think that in a laboratory where it is difficult to make culture, overlooked *L. pneumophila* SG1 infection can easily identified through this method.

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THE EFFECTS OF SULPHASALAZINE ON CELLULAR FUNCTION AND SIGNALING SYSTEM IN T CELL RESPONSE

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The molecular effect of sulphasalazine, which is used in the treatment of rheumatoid arthritis, Crohn's Disease and ulcerative colitis and whose therapeutic effects is accepted to originate from its antiinflammatory and immunosuppressive abilities, is not known in detail. T lymphocytes enter in the S phase of the cell cycle by activation as a response to an antigen. Thus we examined the effect of protein kinase-C (PKC) activation, Ca⁺⁺-Calmoduline (CaM)-dependent activated systems, tyrosine phosphorylation, and Sulphasalazine on the Phytohemagglutinine (PHA), Concalavaline-A (ConA), aCD3+PMA, Mixed Lymphocyte (MLR), and PKC dependent T-Cells responses by using H-7 as the PKC inhibitor, W-7 as the CaM blocker, and genistein (Gen) as the tyrosine kinase inhibitor, in the accessory cell-dependent cell culture systems with and without PKC depleted T-Cells. As a result;

1. In the absence of antigenic/mitogenic stimuli, the viabilities of the dormant phase cells in the culture systems decrease in proportion with the concentration and contact duration of sulphasalazine in the medium.
2. Naturally, the importance of PKC in the signalling system in the basis of cell proliferations depends mostly on aCD3-PMA, together with ConA and PHA in decreasing order.
3. The importance of CaM in the signalling system in the basis of cell proliferations depends on ConA, PHA on aCD3+PMA dependent stimuli in decreasing order.
4. In the importance of PKC in the signalling cascade triggered for proliferative response, primarily aCD3-PMA stimulus, then ConA and PHA stimuli takes place.
5. Of the antigenic/mitogenic stimulators that we used, PHA triggered the proliferation of the PKC depleted cell at the highest level. For proliferation in these type of cells, when more than one signalling mechanism is stimulated, mitogenic stimulators are always superior to antigenic stimulators.
6. The proliferative response for T-Cell specific PHA stimulus inhibites at a desired level even in lowest doses of sulphasalazine concentrations.
7. PKC, PTK and CaM are not sulphasalazine's inner cell target. The inhibition of cellular PKC, PTK and CaM indicate additive effect in sulphasalazine inhibitor.
8. The highest values in the IL-2r expression study was obtained by PHA stimulus. The existence of Sulphasalazine in the medium suppressed this expression by 85%.

RESEARCH ON THE ACTIVE CONSTITUENTS OF SYMPHYTUM SYLVATICUM BOISS. SUBSP. SEPULCRALE (BOISS & BAL.) GREUTER & BURDET VAR. SEPULCRALE

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Symphytum sylvaticum Boiss. subsp. *sepulcrale* (Boiss. & Bal.) Greuter & Burdet var. *sepulcrale* is an endemic Anatolian species. In this study; two pyrrolizidine alkaloids were isolated from *Symphytum sylvaticum* Boiss. subsp. *sepulcrale* (Boiss. & Bal.) Greuter & Burdet var. *sepulcrale*; **Echimidine-N-Oxide (ENO)** from root and **Echiumine** from aerial parts. Also; **Echimidine** was isolated from *Symphytum aintabicum* Hub. - Mor. & Wickens. The structure of the isolated compounds were elucidated based on UV, IR, MS, ¹H and ¹³C NMR analysis. **Echiumine** had been isolated for the first time from *Symphytum* species.

ENO has been determined as the major alkaloid of the roots of *Symphytum sylvaticum* Boiss. subsp. *sepulcrale* (Boiss & Bal.) Greuter & Burdet var. *sepulcrale*. Quantitative analysis of ENO has been performed by GC in roots and aerial parts of *Symphytum sylvaticum* Boiss. subsp. *sepulcrale* (Boiss. & Bal.) Greuter & Burdet var. *sepulcrale*. Crude alkaloid levels range from 0.190-0.208 % in roots and 0.091 - 0.095 % in aerial part. The roots contain 0.1085 % ENO and the aerial parts contain 0.0061 % ENO.

The healing action of the roots and aerial parts of *Symphytum* species may be related to presence of **allantoin**. Quantitative HPLC analysis of **allantoin** in the root, stem - branch and leaves of *Symphytum sylvaticum* Boiss. subsp. *sepulcrale* (Boiss. & Bal.) Greuter & Burdet var. *sepulcrale* were made. As a result; the amount of **allantoin** was found to be in the roots 0.4568%, in the stem-branch 0.4118% and in the leaves 0.1883%.

Toxicity of Pyrrolizidine alkaloids have similar principal symptoms of chronic Cu poisoning. Exposure to pyrrolizidine alkaloids results in high concentrations of liver Cu. According to our opinion; the amount of zinc in *Symphytum* species may be important for their dermatological activities. From the roots of *Symphytum sylvaticum* Boiss. subsp. *sepulcrale* (Boiss. & Bal.) Greuter & Burdet var. *sepulcrale* 5.08% ash and from the aerial parts 11.67 % ash have been obtained. The ash has been analysed and 177.6 µg/g Cu and 341.4 µg/g Zn are determined in the roots and 79.7 µg/g Cu and 200.1 µg/g Zn are determined in the aerial parts.

Antifungal studies on *Heliotropium* species have been a model for the antifungal activities on *Symphytum* species. Antifungal activity is investigated on different extracts of *Symphytum sylvaticum* Boiss. subsp. *sepulcrale* (Boiss. & Bal.) Greuter & Burdet var. *sepulcrale* and ENO. The experiments done with 10 different fungus showed us that the Root Alkaloid Fraction is more effective than the other extracts. We determined that ENO is the compound responsible for the antifungal activity.