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### THE PAST AND THE PRESENT OF CITRUS CERTIFICATION IN CROATIA

# D. Škorić, M. Krajačić

Department of Biology (Botany), Faculty of Science, University of Zagreb, Zagreb - Croatia

#### D. Hartl, Ž. Gatin

Institute for Adriatic Crops and Karst Reclamation, Split - Croatia

**SUMMARY -** The main characteristics of the citrus industry in Croatia are given. The sanitary status is outlined as well as the perspectives for the establishment of the citrus certification program.

**Key words**: Citrus, certification, viruses, virus-like pathogens, Croatia

**RESUME -** Les caractéristiques majeures de l'agrumiculture en Croatie sont données, L'état sanitaire est mis en évidence de même que les perspectives d'établissement d'un programme de certification des agrumes.

Mots clés: Agrumes, certification, virus, pathogènes de type viral, Croatie

### THE BEGINNINGS OF A MODERN CITRUS PRODUCTION

The development of the modern citrus industry in Croatia started about 40 years ago as an integral part of an FAO project of land reclamation in the Lower Neretva Valley. The Neretva Project, UNDP/SF/FAO. No. 63 became operational in May 1963. To realize such a project, it was necessary to create a suitable cultivar program. With the help of experts engaged by FAO, L. Blondel, H. Chapot, A. D. Alexandrov, E. Calavan, C. N. Roistacher and others, more than 110 types of citrus were collected from Corsica, California, Georgia and Japan. The citrus foundation block was established in the period of 1964 - 1984 at the Experimental field in Neretva Valley. Unfortunately, the complete foundation citrus stock - species, cultivars and clones, collected and established in the Experimental Field Station in Neretva Valley from 1964 onwards, was lost at the time of war (1991-1995) in ex-Yugoslavia (Gatin, 1992). Today, the rebuilding of the citrus foundation stock is an indispensable condition for further development of the citrus industry in Croatia.

#### CITRUS VARIETIES AND PRODUCTION CONSTRAINTS

Croatia grows citrus in Dalmatia, the southern coastal region with the islands, which is between 42° and 44° of the northern latitudes and represents probably the northernmost commercial citrus growing area in the world with the climatic conditions comparable probably only to the Sukhumi region in the ex-USSR republic of Georgia. Therefore, the cultivar program is mostly limited to the cold tolerant Satsuma mandarin (*Citrus unshiu* Marc.), but the lemon and the sweet orange can be grown in some warmer micro-locations (e.g. the lemon groves on the island of Vis). The early ripening season of Satsuma cultivars is another important factor for avoiding the potential winter freezing of the fruits. This type of citrus is well accepted by the Croatian market, as well as by the markets of the neighboring countries. It faces less competition from the big citrus importers, especially the very early and early-ripening Satsuma cultivars like Zorica Rana (obtained by a bud mutation from a Japanese early cultivar Kawano Wase) and Wakiyama (Gatin, 1997). For all the reasons mentioned above, ever since the introduction of early Satsuma cultivars, producing them has been a lucrative family business.

Compared to the biggest, our citrus industry with the acreage of 1500 ha and the production of up to 30.000 tons of Satsuma mandarins per year, is tiny but still important for the local economy.

An average Croatian citrus producer is consequentially strongly oriented towards the very early and early-ripening Satsuma cultivars and the demand for the propagating material is increasing. It is estimated that in the last five years 2.5 millions of various Satsuma mandarin nursery trees have been produced (about 500,000/year), but unfortunately almost all of them were produced out of the certification program. The need for the propagating material is likely to drop after this initial post-war expansion, but still some 200,000 nursery trees will probably be needed yearly.

#### **SANITARY STATUS OF CROATIAN CITRUS**

The citrus growing in coastal Dalmatia and on the islands is known since the 15th century. Citron, lemon, sour orange and sweet orange, later, were introduced from Sicily, Portugal and other Mediterranean countries by sailors. For centuries, they were grown only within the boundaries of the family yards. At first, they all came as seedlings and they were propagated by seeds. Sour orange as a rootstock was not used until late 19th century. The first citrus nursery was founded in Čibača near Dubrovnik in 1908. Owari Satsuma on *Poncirus trifoliata* L. Raf. rootstock was introduced from Japan in 1933/34, the Navel oranges from the USA in 1932 and 1935, and Moro and Tarocco oranges from Italy in 1937. The systematic work on the introduction and evaluation of citrus began in 1937 and it was performed by the Institute for Adriatic Crops in Split with its experimental stations in Dubrovnik and Topolica (now Montenegro). After the introduction of P. trifoliata as a rootstock, it became clear by the bark cracking symptoms on the rootstock that the cultivars, which had been grown in Dalmatia before the establishment of the citrus foundation block, were contaminated by Citrus exocortis (CEVd) and possibly other Citrus viroids (CVds). Only Satsumas remained symptomless. The first Washington navel and other sweet oranges on sour orange rootstock were probably free of Citrus tristeza closterovirus (CTV). Unaware of the potential dangers caused by CEVd and other Citrus viroids, in the following years most of the propagations were done on *P. trifoliata* rootstock.

We have a reason to believe that the CTV arrived in the country with the Satsuma mandarins introduced as early as 1933-1934 from Japan. The first report of CTV in ex-Yugoslavia (presumably Croatia) was by Italian authors Davino and Catara (1986) who detected CTV in Satsuma mandarin budsticks. This material had been introduced into the Croatian nurseries in 1980's from Japan (Šarić and Dulić, 1990). Most of the Croatian Satsuma trees stem from this material; over 90% is grafted on *P. trifoliata* and shows no symptoms. With *P. trifoliata* being the main rootstock in this area and cultivation of many symptomless citrus varieties, CTV has probably spread but without causing strong field symptoms or major production losses. Still, having in mind potential danger, the trees of the old foundation block were tested regularly to CTV by bioindicators and ELISA (Šarić and Dulić, 1990) and "new introductions" from the late 1970's were done preferentially from virus-free stocks. Some examples being: Saygon SRA-29 and Owari SRA-145 Satsuma mandarins from Corsica, Clementine SRA-63 and 64, Lisbon and Eureka lemons, Red Blush grapefruit and other cultivars from Corsica, as well as Meyer lemon and Skagg's Bonanza Navel from Willits & Newcomb Nursery.

The assessment of the present sanitary status of Croatian citrus has been under way as an integral part of the projects "The Introduction of Citrus in the Dalmatian Coastal and Island Area" and "Plant Viruses, Subviral Pathogens and Phytoplasmas" financed by the Croatian Ministry of Agriculture and Forestry and the Croatian Ministry of Science and Technology, respectively. Considering the importance, geographical distribution and the specific characteristics of our citrus production, the stress has been put, so far, on the detection and diagnosis of CTV and Citrus viroids by using biological indexing, dsRNA, sPAGE, RT-PCR, and other relevant molecular biology methods as one of the first steps within the prospective of the citrus certification scheme. Different CTV strains have been detected (Černi et al., 2002) and, besides CEVd, some other Citrus viroids (CVd-II, CVd-III) are found to be a part of the viroid complexes in the Croatian citrus (Škorić, 2000; Škorić et al., 2001). The detection of Citrus psorosis virus and Satsuma dwarf virus by biological indexing and ELISA are soon to be introduced. Hence, the assessment of the Croatian citrus sanitary status is at the very beginning. The number of samples taken is far from being exhaustive and the results are still not informative enough to have a realistic picture of the present sanitary status in the collection groves and nurseries.

#### EFFORTS TOWARDS ESTABLISHING THE CITRUS CERTIFICATION PROGRAM

The establishment of the new citrus groves in the post-war period, the loss of the preexisting foundation block and the new Croatian legislative measures prompted us to take decisive steps towards the establishment of the citrus certification program. The Croatian Parliament passed a law (NN131/97, *The Bill on the seeds, reproduction material and varietal acknowledgement of the cultivated plants*) which from the year 2007 onwards, requires the introduction and production of certified plant material only. This is the way to improve the citrus growing in Croatia and adjust it to the EU standards, as well as to bring quality products to the domestic and global market.

The first attempt towards organizing financial support by the international sources was done in March 1998, when Enrique de J. Arias-Jimenez, from FAO Plant Production and Protection Division, visited the Institute for Adriatic Crops and Karst Reclamation-Split, one of the institutions actively engaged in this project. Mr. Arias-Jimenez and the scientists at the Institute (Slavko Perica and Živko Gatin) made a draft of a small project concerning citrus collection and organization of a foundation block, which was unfortunately never realized due to the lack of funds and budget cuts in Croatia. However, the political will exists and the awareness for funding the citrus certification has been raised especially in the local community. The first tangible result was a joint financing of the greenhouse for the establishing citrus foundation block and certification purposes from the local government of the Split-Dalmatian County (about 13 000 USD) and the Croatian Ministry of Agriculture and Forestry (50 000 USD). Although we have scientific and technical staff, as well as enthusiasm, the lack of funds from domestic as well as from the international sources is still a major problem.

Several projects with the aim to establishing the citrus foundation block, a grove of selected high-quality citrus trees consisting of virus and viroid-free plants with the special attention to the cultivation of (Satsuma) mandarin, are proposed to the different bodies of the Croatian goverment. A set of technological procedures and measures will be applied encompassing biological and molecular detection of virus and virus-like pathogens performed by a team affiliated to the Department of Biology (Botany), Faculty of Science, University of Zagreb (Silvija Černi, Mladen Krajačić, Dijana Škorić). Thermotherapy, shoot-tip grafting, and the establishment of the increase block and the foundation block itself will take place at the Institute for Adriatic Crops and Karst Reclamation-Split led by a team of three scientists (Živko Gatin, Danijela Hartl, Katarina Hančević). The cooperation with other certification programs and the harmonization of protocols within the MNCC respecting all the technical and scientific guidelines is expected to help the production of high-quality citrus propagating material in Croatia, in both sanitary and pomological aspect, through easier exchange of relevant information.

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