EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION

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Report of the 5th Meeting of the EPPO ad hoc Panel on *Diabrotica virgifera virgifera* held jointly with the 7th International IWGO Workshop on *Diabrotica virgifera virgifera*

Stuttgart, DE, 2000-11-16-17

1. Opening

The participants (see Appendix I) were welcomed by. Dr Buchenauer and Dr Boehringer of the University of Hohenheim, Stuttgart (Germany) and by Dr Berger and Prof. Edwards on behalf of IWGO.

2. Situation of *Diabrotica virgifera virgifera* in the EPPO region

The situation of *D. virgifera virgifera* has been reviewed and several papers presented the current spread of the pest in Europe. In summary, the pest has continued to spread in 2000. It has been found for the first time in 2000 in Slovakia. Looking at a physical map of this region, it seems that *D. virgifera* has almost filled up what could be called the great Hungarian plain. But surprisingly, it has also been caught outside this region. Similarly to the previous finding near Venezia airport, *D. virgifera* has unexpectedly been found near 2 new airports in Italy and Switzerland: in Milano (Malpensa) and Lugano/Agno airports. The map in Figure 1 shows the spread of *D. virgifera* in Europe from 1992 to 2000. Economic damage has been seen on maize in Serbia, and also in some bordering areas in Croatia, Hungary and Romania. The map in Figure 2 shows the areas in which economic damage has been observed. As a general remark, Prof. Maceljski felt that countries should intensify their trapping programmes and that more international cooperation is needed.

Albania

Monitoring started in Albania in 1999 reported Dr Çota. In 2000, pheromone and yellow sticky traps were placed in maize fields in 7 counties (Shokodra, Bushat, Elbasan, Peshkopi, Durres, Sarande and Lushnja) and near the Rinas International Airport. A specimen looking like *D. virgifera* was found near the International Rinas Airport but was shown after detailed examination to be another species. In conclusion, *D. virgifera* has not yet been found in Albania.

Austria

Dr Berger reported that pheromone traps were placed along the borders with Slovenia, Hungary and Slovakia and near Vienna airport. In Austria, Steiermark is probably the most endangered region, as maize and pumpkins are widely grown. No *D. virgifera* was found in 2000. In 2001, monitoring will be intensified and conducted on larger areas.

Bosnia & Herzegovina

D. virgifera was first found in 1997 in areas bordering Serbia and Croatia. Mr Berberovic presented the results of the monitoring programme carried out in the Federation of Bosnia & Herzegovina. 71 monitoring sites (pheromone and yellow sticky traps) were placed within the infested area and in endangered places where spread of the pest was expected. Traps were placed in Posavina, Tuzla, Zenica-Doboj and Una-Sana cantons. A total of 4374 beetles was caught in Posavina, Tuzla, Zenica- and Doboj cantons. No beetles were trapped in Una-Sana canton. Most catches were made in Posavina and Tuzla cantons. In 2000, the spread of *D. virgifera* to new areas was limited (increase of 150 km²

compared to 1999) but population levels have greatly increased. It is now estimated that the infestation covers 12,000 km², corresponding to 70% of the maize-growing area. No economic damage is reported from the Federation of Bosnia & Herzegovina.
Prof. Baca presented the 2000 situation for Republica Srpska. Results of the monitoring programme showed that spread continued towards the west (now approximately 150 km away from the Yugoslavian border). As the summer was very dry and hot, adult populations were active earlier than in previous years.

Bulgaria
Prof. Edwards presented the report prepared by Ms Ivanova. A monitoring programme was initiated in Bulgaria in 1995. The first *D. virgifera* beetles were caught in 1998 in the north-west, near the Serbian and Romanian borders. In 2000, monitoring started at the beginning of July. Pheromone and yellow sticky traps were placed in the north and northwest of Bulgaria, near the cities of Vidin, Vratsa, Montana, Pleven, Veliko, Tarnovo and Sofia. The highest numbers of beetles were caught near Bregovo (372 adults), close to the Serbian border and in Gramada (214), Prevala (290), Mitrovei (138). In 2000, *D. virgifera* was also found in the region of Dimova and Montana. The conclusion was that the pest has continued to spread within Bulgaria.

Croatia
D. virgifera was first found in the east of Croatia in 1995, and has then spread towards the west of the country. Prof. Maceljski presented the results of the 2000 monitoring programme. 130 trapping sites with pheromone and yellow sticky traps were monitored. In total, 15,084 beetles were caught in 2000. It was estimated that the infested area has reached 14,500 km² (on which 200,000 ha of maize are grown). The first beetles were caught 10-14 days earlier than in previous years (first catch on 20th June) because of the extremely dry and hot weather conditions prevailing in May and June. The pest continued to spread towards the west in the northern and middle part of the infested area (35 km in the north, 20 km in the middle part). But no spread was observed in the south of the infested area. The farthest point of spread towards the west is approximately 40 km away from Slovenia. Damage was observed only in some fields and was also related to the very dry conditions. In average, good results were obtained with chemical treatments (band treatment applied at sowing).

Czechia
In 2000, 34 monitoring sites were put in place in South Moravia and in the vicinity of airports (Prague and Ostrava) and were checked from June to beginning of October. *D. virgifera* was not found in Czechia.

Germany
Monitoring programmes have been carried out in Germany since 1998. Mr Botzenhardt presented in detail the monitoring programme followed in Baden-Württemberg which is considered as a high risk area for *D. virgifera* in Germany. According to the present situation of the pest in Eastern and Central Europe, it is felt that it will continue to spread naturally along the Danube and will enter Germany first in Bayern and then in Baden-Württemberg. However, the pest can also be transported by other means. Therefore, in 2000, pheromone and MCA (plant kairomone) traps were placed in 30 locations in Baden-Württemberg in maize-growing areas, mainly along motorways, near airports, military installations and near places where large quantities of commodities are handled. No *D. virgifera* were found in Baden-Württemberg nor elsewhere in Germany.

Hungary
Mr Ripka presented the situation in Hungary in 2000. *D. virgifera* was first found in Hungary in 1995, in the south of the country and has then spread very significantly. In 2000, monitoring continued with pheromone and yellow sticky traps. The pest was trapped in 16 out of the 19 Hungarian counties. It is now present in the counties of Borsod-Abaúj-Zemplén, Nográd, Veszprém and Zala. It continued to spread towards the north, in particular along the river Tisza, and it has reached the Slovakian border. Towards the west, it has also reached the northern shore of lake Balaton. As in previous years, the highest numbers of adults were caught in the south of the country. Larval damage was seen in the

following counties: Békés, Baranya, Bács-Kiskun and Csongrád on 3,103 ha. Economic damage was observed in some areas in Bács-Kiskun and Csongrád counties.

Italy

Dr Furlan and Vettorazzo presented the measures taken in the Veneto region to eradicate *D. virgifera.* Following the introduction of *D. virgifera* in Yugoslavia, a monitoring programme was put in place in Italy in 1995. In summer 1998, 7 specimens of *D. virgifera* were caught for the first time in one locality Tessera near the Marco Polo International Airport of Venezia. In 1999, 2 specimens were caught near the airport. Since 1999, phytosanitary measures have been taken to eliminate the pest and prevent any further spread. A Ministerial decree on compulsory control against *D. virgifera* is now in force in Italy.

In 2000, the containment and eradication measures implemented were the following:

1) Monitoring of *D. virgifera* populations in the quarantine area (1200 ha) and a surrounding buffer zone (within a radius of 22-25 km, i.e. 35,000 ha), using pheromone traps in maize fields (400 traps with a 400 x 400 grid in the quarantine area, and 250 traps in the buffer zone with a 1 x 1 km grid). Some additional traps of various types were placed for research purposes in the quarantine area.
2) Maize monoculture was not allowed.
3) Insecticide treatments[1] against the adults were applied twice (from July to end of August) in all maize fields throughout the regulated area (quarantine area and buffer zone).
4) Movement of fresh maize or soil in which maize was grown during the previous year was prohibited from the quarantine area.
5) Harvest was not allowed before the 1st October.

This containment and eradication programme was financed by Veneto region and EU. The total cost in 2000 was estimated at approximately 81,500 EUR.

In the quarantine area, all fields which had been planted with maize in 1999 were inspected in 2000 to see which crop was grown. As a result, 6.7 ha were recorded in June as maize monoculture (maize was cultivated in 1999) and these fields were immediately sprayed and destroyed. This was difficult to achieve in practice, as farmers saw no necessity to destroy their healthy looking fields. Despite these efforts, 4 very small plots of maize planted for home consumption (0.3 ha) were identified later in July, hidden between houses and vegetable crops. Maize had been grown on these plots for the last 4 years. 70 pheromone traps were placed in and around these plots. 73 *D. virgifera* males were caught in this small area from 26 to 29 July 2000. Insecticide treatment was applied on the 29th July. 2 more specimens were caught during the 1 or 2 days following this treatment. The insecticide application was repeated 7 days after and since then no other specimens were captured. No *D. virgifera* was caught in the surrounding zone.

It was concluded that the area in which insects can be caught has been drastically reduced from 1998 from 2000 (while the trapping intensity has increased). The key factor in eradicating the pest appears to be the suppression of maize monoculture. However, it was stressed that the existence of very small areas of monoculture is sufficient to maintain rather high populations of *D. virgifera*, and therefore to ensure reproduction and spread of the pest. It was also noted that insecticide treatments against adults were efficient and stopped their spread.

Other regions in Italy have started to monitor for *D. virgifera*. Surprisingly, 3 specimens of *D. virgifera* were caught for the first time in Lombardia, near Milano International airport (Malpensa). No further details were provided during the meeting on this new finding.

Romania

The first find of *D. virgifera* was made in 1996 at Nadlac (Arad district bordering Hungary). In the following years, the pest has spread towards the east (Figure 1). According to a report prepared by Mr Vonica, *D. virgifera* was caught for the first time in 2000 in the following counties: Satu Mare, Salaj, Alba, Gorj and Olt. Compared to previous years, population levels have increased in 2000, especially in Caras-Severin, Timis, Arad, and Mehedinti counties. Larval damage was noted in 1999 in maize

[1] Chlorpyrifos (Dursban WG, 1.1 kg/ha) the only registered product against *D. virgifera* in Italy.

monoculture in some areas in the Caras-Severin county, and economic damage appeared in 2000 in these areas. Because of high temperatures prevailing in spring 2000, the first adults appeared approximately 3 weeks earlier than in previous years. Mr Rosca presented the potential strategies which could be used in Romania to control *D. virgifera*. Studies have showed that there is no climatic limiting factor to prevent the spread of *D. virgifera* in Romania to all maize-growing regions. Rotation of maize with other crops appeared as the primary management strategy. However, in Romania maize is grown over 3 million hectares and often as a monoculture. Seed treatments which are currently applied against *Tanymecus dilaticollis* and *Agriotes* species in Romania are not efficient against *D. virgifera*. Mr Rosca felt that, in Romania, control of *D. virgifera* will have to rely on the application of granular insecticides during cultivation and possibly on the use of genetically modified maize.

Slovakia
Monitoring programmes have been conducted in Slovakia since 1996. In 1999, recalled Mr Siviček, *D. virgifera* was already very close to Slovakia. In 2000, pheromone and yellow sticky traps were placed along the borders with Hungary and Austria, as well as near the airports of Bratislava and Košice. The first adult of *D. virgifera* was caught in a pheromone trap in the district Vel'ký Krtíš on the 7th July. 11 other adults were then caught in Vel'ký Krtíš district again and also in Komárno and Lučenec districts (all located in the south of the country). This is the first report of *D. virgifera* in Slovakia.

Slovenia
As in previous years, reported Mr Pajmon, a monitoring programme was carried out in Slovenia. Pheromone and yellow sticky traps were placed at 50 locations mainly along the Croatian and Hungarian borders as this is an important maize-growing area. Additional traps were also placed near the Italian border and the International Airport in Ljubljana. In 2000, no *D. virgifera* was found in Slovenia.

Switzerland
In 2000, *D. virgifera* was reported for the first time in Switzerland, near the airport of Lugano/Agno, in Ticino. In July 2000, 4 adults were caught in 4 traps. As a result of this new introduction, measures were taken. Information on maize crops located within a radius of 5 km around the finding location was collected. All machinery used for harvest was disinfested. In this area, maize production will be prohibited (this concerns 8 farmers). The authorities also gathered information on flights from Eastern and Central European countries which arrived in 1999 and 2000 to try to find the possible origin of this introduction. In 2001, a larger and more intensive monitoring programme will be carried out in Switzerland.

Ukraine
Ukraine is an important maize producer, reported Dr Melnyk. In 2000, this crop has been sown over 1,300,000 ha mainly in the southwestern and southern parts of the country. As it is expected that *D. virgifera* will continue its spread towards Ukraine, pheromone traps have been placed along the Hungarian and Romanian borders. In 2000, no *D. virgifera* was found in Ukraine. Research is currently being done on the efficiency of various types of traps and on the resistance of maize hybrid cultivars to *D. virgifera*.

Yugoslavia
Dr Sivcev recalled that *D. virgifera* was first found in Europe near Belgrade airport in 1992. Monitoring continued as in previous years (300 pheromone traps were placed in 10 counties). No more movement of the pest towards the south was observed, as it is a region of high mountains. Spread continued towards the Southeast, in the region of Negotin. Economic damage was observed on a significantly larger territory (Figure 2). It is estimated that 67,550 km² were infested by *D. virgifera* and that damage occurred on 50,000 ha of maize fields (the area has almost doubled compared to last year). In 2000, damage was rather severe also because the season had been extremely dry. It is expected that the area of economic damage will continue to increase in coming years. However, in areas where damage has been seen for several years, the level of damage is decreasing because maize is now rotated with other crops. Dr Sivcev also reported that 300 ha of hybrid seed maize planted after

soybean were damaged. Further monitoring will be made in this area to study this situation. It can be recalled that similar situations have been observed in some parts of USA (Indiana, Illinois, Michigan, Ohio). It was suggested that this is caused by the emergence of variant populations of *D. virgifera* which have adapted to crop rotation by laying eggs in soybean fields. According to these preliminary observations made in Serbia, it seems that these variant populations also exist in Europe.

4. Research reports

Prof. Edwards presented a paper on the interaction and impact of soil properties on *D. virgifera*. Many soil factors have an impact on survival and behaviour of larvae. High percentage of clay will favour larval survival as well as high moisture (but not saturation). Sandy soils and dry soils are not very favourable to rootworms. Fine texture soils will facilitate movement of larvae in search of food on longer distances. Carbon dioxide is also an important factor which triggers rootworm movement towards maize roots.

Dr Baufeld presented a paper on possible containment and eradication measures against *D. virgifera*. He recalled that according to Council Directive 2000/29/EC, all EU member states should prohibit the introduction of listed *Diabrotica* species (Article 3) and notify any finds of the pest (Article 16). According to ISPM no. 9 (Guidelines for pest eradication programmes), the eradication process involves three main activities: surveillance, containment, and treatment and/or control measures. To investigate the distribution of the pest, countries should focus on the most endangered areas (e.g. maize monoculture areas, important maize-growing regions) and potential points of entry (e.g. airports, sea ports, railways, military installations, breeding stations). It was also felt that cucurbitaceous crops should be included in monitoring programmes. Before a possible introduction of the pest, growers and companies involved in maize production should be informed of this threat. Monitoring should be put in place as soon as possible, so that introduction (if any) is detected as early as possible. Containment and eradication measures can include the following aspects: mass trapping, chemical treatments against larvae and/or adults, prohibition of maize growing in infested fields, changes in rotation, destruction of maize volunteer plants, use of tolerant/resistant/genetically modified cultivars. It was concluded that early detection of the pest and fast reaction was decisive for successful eradication.

Several papers were presented on traps. Dr Wilde presented new types of lure traps (e.g. baited with MCA: 4-methoxy-cinnamaldehyde) which could be used to catch *D. virgifera*. Mrs Ivezic presented the first results of comparison studies done in Croatia using yellow sticky traps and lure traps (feeding attractant + carbaryl). Preliminary results showed that lure traps gave good results. Dr M. Tóth presented a paper on trapping methods and effective trapping range for *D. virgifera*, and in particular on the use of non-saturable, high capacity trap design which could be used for quantitative monitoring of pest populations.

Dr Tollefson presented the new Iowa scale used to assess larval root damage. This scale is based on the number of full nodes eaten (X) and percentage of a node eaten (YY). This scale ranges from no feeding damage (0.00) to 3 or more nodes eaten (3.00). As a example, if more than one node has been eaten and the additional injury is equivalent to half a node, the rating would be 1.50. The estimated economic injury level is 0.25 (a quarter of a node destroyed).It is felt that this new linear scale is more intuitive and more precise than the previously used Iowa scale (non-linear scale rating from 1 to 6). The new scale can be viewed on Internet:
http://www.ent.iastate.edu/pest/rootworm/nodeinjury/nodeinjury.html.

Several papers were presented on the biology of *D. virgifera*. Ms Zseller observed larval development, adult emergence of *D. virgifera* in the field, in the south of Hungary. Mr Möser presented preliminary results on the food resources used by adults of *D. virgifera* in southern Hungary. Diet composition followed maize phenology and also the diversity and abundance of weeds inside maize fields (such as: *Amaranthus*, *Echinochloa crus-galli*, *Sorghum halepense*) or remaining plants of the previous crops

(such as: *Cucurbita maxima*, *Helianthus annuus*). Mr Töpfer presented the first results of a study on life table parameters done in the field (in Hungary) and in the laboratory. Mrs Mucsi presented a study on the population structure of *D. virgifera* observed in Szeged (south of Hungary).

Prof. Kiss presented a study on crop rotation to control *D. virgifera*. A three-year crop rotation trial has been set up in 2000 in Szeged (south of Hungary). Crops included were: maize, sunflower, soybean and a cereal (oat or winter wheat). Preliminary results showed that adults emerged only in plots where maize was grown after maize. However, final results of this study will be presented at the end of the three-year study.

Several papers were presented on chemical control of *D. virgifera*. Dr Sivcev presented the result of an efficacy trial with 8 insecticides currently registered in Yugoslavia for seed treatments of maize. Preliminary results showed that tefluthin and imidacloprid gave the best results, but damage could still be observed in some roots. He felt that seed treatments may not always give satisfactory results depending on the time of planting and emergence of first larvae. Mrs Mucsi presented a study on the penetration of aerial sprays within maize canopy. Mrs Keresi presented a study on the efficacy of several insecticides done in Yugoslavia.

Dr Wennemann presented a paper on the potential use of MCA as an orientation disruption tool against *D. virgifera*. Preliminary analysis of field trials showed some variable orientation disruption of the adults. Further studies on orientation disruption will continue in 2001.

Dr Kuhlmann presented the potential biological agents which could be used against *D. virgifera*. Research is currently being done in collaboration with scientists from Central and South America to try to identify natural enemies which could then be introduced and released in Europe. So far, Tachinids (e.g. *Celatoria* species) and Braconids (e.g. *Centites* species) have been identified, but further studies on their biology, ecology, efficacy and potential non-target risks are needed.

Dr F. Tóth presented a study on the potential impact of a predator spider species (*Enoplognatha latimana*) on the silk damage caused by adults of *D. virgifera*.

Finally a presentation of a genetically modified maize (MaxGard) resistant to *D. virgifera* was made to the participants. A *Bacillus thuringiensis* protein (Cry3Bb) specific to chrysomelids is synthesized by this cultivar. It is expected that this new resistant hybrid cultivar will be launched in 2002, in USA.

For more information, all abstracts of the papers presented during this meeting can be viewed on the IWGO Web site: http://www.infoland.at/iwgo/

5. Close

On behalf of the participants, Dr Berger, Prof. Edwards and Ms Roy (EPPO) warmly thanked Dr Boehringer and his colleagues for the fine organization of this meeting. The next joint meeting will take place in Venezia, Italy on the 30th October 2001.

Figure 1
Spread of *Diabrotica virgifera virgifera* in Europe 1992-2000 (by C.R. EDWARDS and J. KISS; based on data from Festic, Furlan, Igrc-Barcic, Ivanova, Maceljski, Princzinger, Romeis, Sivicek, Sivcev and Vonica)

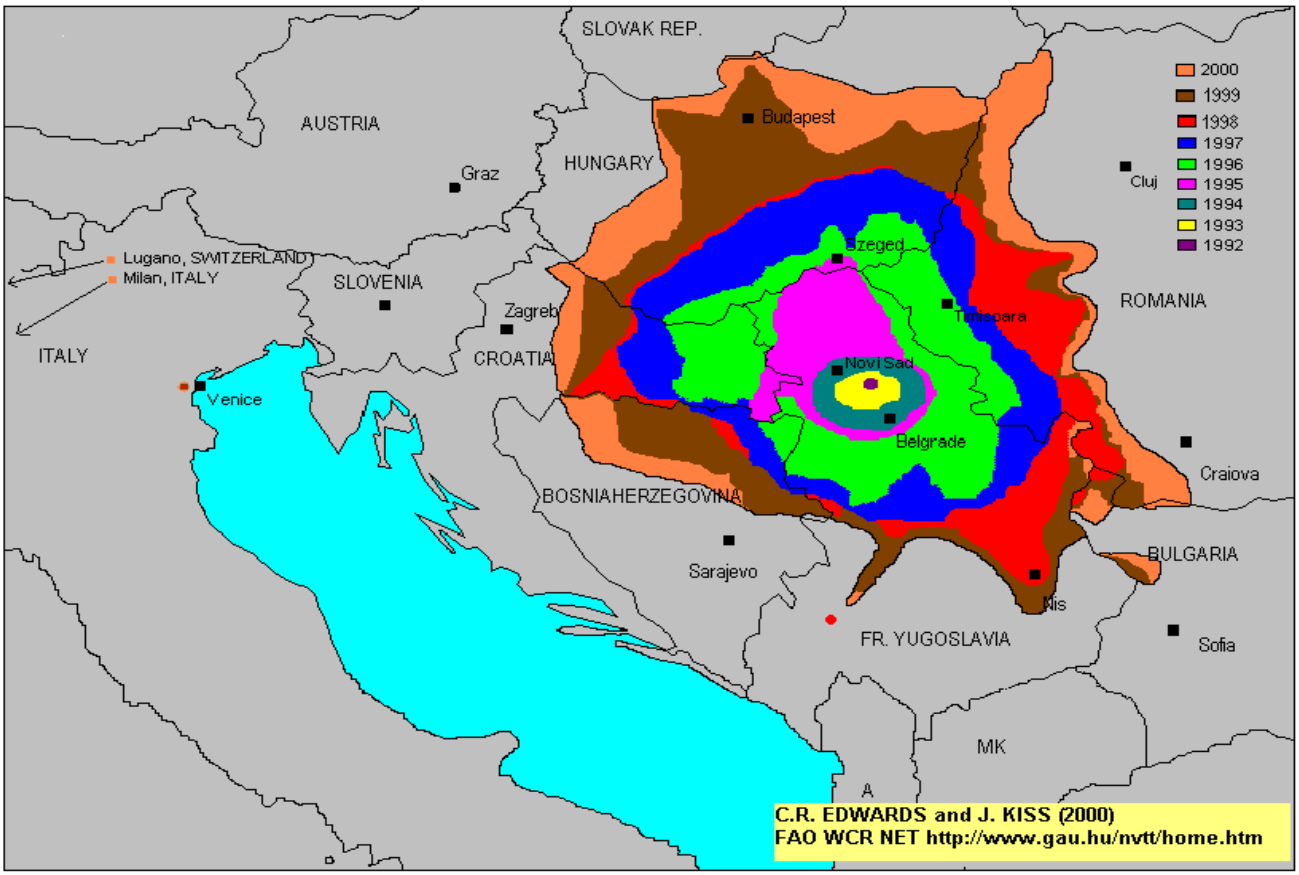
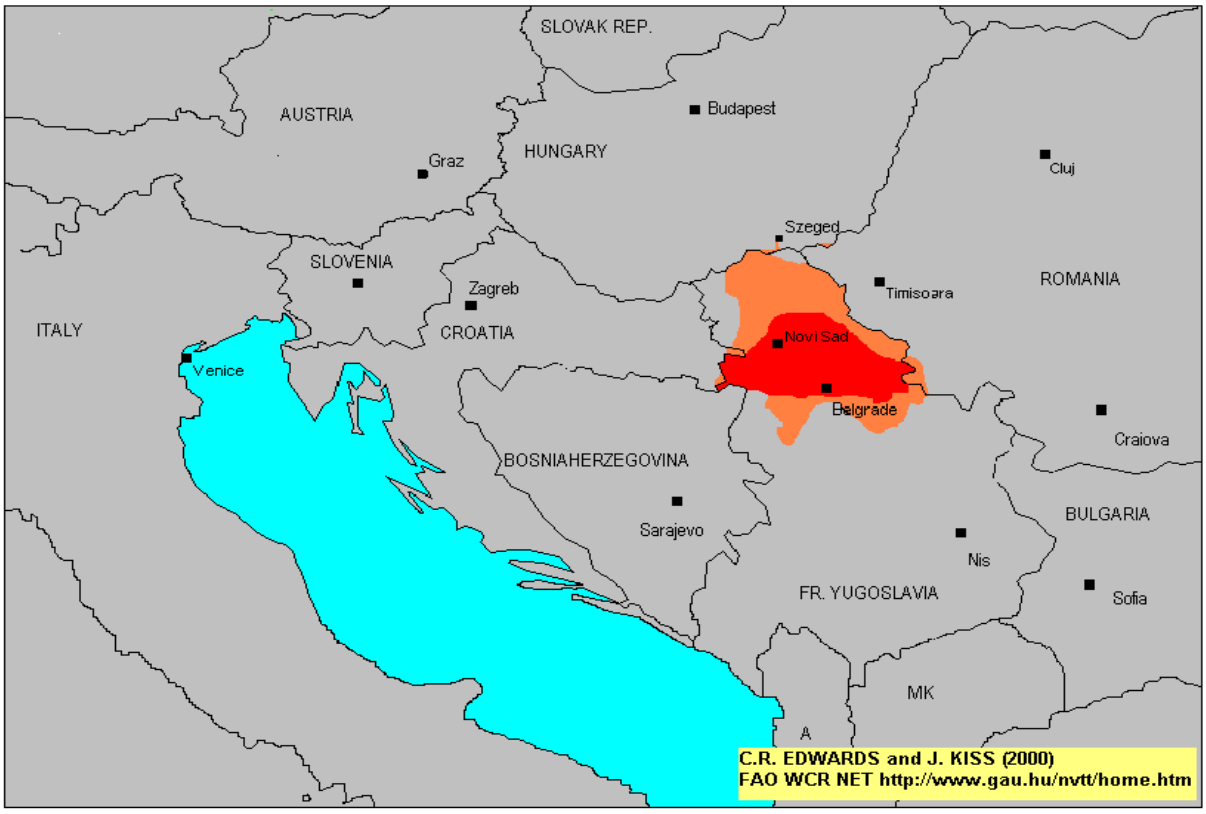


Figure 2
Economic larval damage of *Diabrotica virgifera virgifera* in Europe in 2000 (by C.R. EDWARDS and J. KISS; based on data from Igrc-Barcic, Princzinger, Sivcev and Vonica)



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