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Address of Editorial office:

Agricultural Science and Technology
Faculty of Agriculture, Trakia University
Student's campus, 6000 Stara Zagora
Bulgaria

Telephone.: +359 42 699330
+359 42 699446

<http://www.uni-sz.bg/ascitech/index.html>

Technical Assistance:

Nely Tzvetanova
Telephone.: +359 42 699446
E-mail: ascitech@uni-sz.bg

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Breeding and agrotechnics of rape (*Brassica napus* L.). Winter rape - distribution, cultivation and investigation in Bulgaria

M. Hristova-Cherbadzi^{1*}, G. Georgiev²

¹Agronomy Faculty, University of Forestry, 10 Kl.Ohridski, 1756 Sofia, Bulgaria

²Dobroudja Agricultural Institute, 9520 General Toshevo, Bulgaria

Abstract. Oilseed rape (OSR) or rapeseed - *Brassica napus* L. is the most important plant of genus *Brassica*, grown for its oil content. Breeding of new rape varieties without eruc acid content ("0" type) and low-glucosinolytic content ("00" type) is a prerequisite for the cultivation of this crop in more countries and on increasingly larger areas. The recent gradual re-introduction of rapeseed in our country turned it into a successful crop. The potential of the introduced winter and spring rape varieties was investigated. The technology for new varieties and hybrids grown in Europe and worldwide was studied and improved. New initial material was collected and examined as a basis for the resumed breeding of rapeseed in Bulgaria. The research work was directed towards developing parental forms and using heterosis in rapeseed.

Keywords: *Brassica napus* L., breeding, winter rape, hybrid

Abbreviations: OSR - oilseed rape, CMS-cytoplasmic male sterility, GM-genetically modified

Introduction

Rapeseed (*Brassica napus* L.) together with turnip, radish, shepherd's bag, mustard and others belong to family *Brassicaceae*, genus *Brassica*. According to data presented by FAO in 1998, the world collection of plants from genus *Brassica* (crop: mustard/rapeseed) listed 106 923 accessions and represented one of the biggest germplasm collections in the world. The name of rapeseed comes from the Latin word "rapum" or "rapa", which means turnip. *Brassica napus* is used not only for industrial purposes but also for human consumption and animal food (Erić et al., 2006) (Figure 1).

Winter oilseed rape has high nutrition value and can be used as green fodder. In 100 kg of green mass there are up to 4 kg of protein, or 16 units of beetroot. One unit of beetroot green mass of winter rape accounts for 180-190 g protein. After seed processing the obtained valuable protein meal is suitable for adding to the production of compound feed. Meal from the seeds of varieties without eruc glucosinolates contains minor amounts (upto 0.5 %), and food quality is equivalent to soybean meal, which contains 45-49 % protein with high amino acid content (Staneva et al., 2007). Rapeseed is also early food for bees. Up to 10 kg of honey are obtained per 0.5 ha. It is one of the "energy crops" (Directive 2003/96/EU; Regulation 1782) and the European Union Directive No.30 of 2003 (Directive 2003/30/EU) requires an increase in eco-fuel (biofuels) at the expense of oil derivatives. Even Rudolf Diesel in 1912 mentioned that oil plants could be used for obtaining diesel to replace fuel (Wagener-Lohse, 2006). According to Todorova (2006), Bulgaria has the potential to produce biofuels. The total area of abandoned agricultural lands (based on MAF) during the period from 2002 to 2007 was about 460 000 ha (8 % of all arable land). Institute of Soil Science "Poushkarov", Sofia has developed a geographical information system to aid agricultural producers. It includes the

study of agricultural land in terms of the choice of optimal conditions for cultivation of energy crops used for biofuel production (Banov and Georgiev, 2006).

Rapeseed has deep and fast growing root system which improves soil structure and increases water permeability and aeration. The winter forms have high soil-protection effect. In rice fields, where fertilization with manure is difficult, rapeseed can be used successfully for green manure. It improves plant health and soil, reducing root rot damages in wheat. Used as an intermediate crop, rapeseed is removed first from the fields leaving them free from weeds for the next crop. This helps to increase the effectiveness of the use per unit area. According to Oil World, the world production of rapeseed in 2007 was with an estimated level of 51.6 million tons based on data for 2006 - 47.2 million tons. The main producer of OSR is China, followed by India, Canada, Germany, France and the UK (Marjanović-Jeromela et al., 2008). The development of new double-zero type varieties, construction of refineries for processing rapeseed oil and Bulgaria's joining the EU will increase the demand for rapeseed processing and export. From 1 200 ha in 1997 the areas planted with rapeseed in Bulgaria reached 11 000 ha in 2005 (Staneva et al., 2007). In recent years rapeseed is being grown on increasingly larger arable land (Table 1). It is represented by foreign varieties and hybrids which are characterized with high oleic acid oil.

Oilseed rape is the most important plant of genus *Brassica*, grown for oil. Among the oil seed crops grown worldwide, rapeseed ranks third after soy and palm. The average oil content in the seeds of the varieties is 41 to 44 % and the average seed yield depending on the specific agroclimatic conditions and production in Bulgaria is 1500 to 3000 kg/ha.

The nutritional value of rapeseed oil is determined by the fatty-acid composition and it contains biologically active substances (vitamins A, D, E and K), phosphatides, tocopherols, etc. The valuable essential fatty acids are about 85 % of the oil composition: oleic (65 %) and linoleic (20 %). Such high oleic acid oil has high

* e-mail: mirahristova@yahoo.com

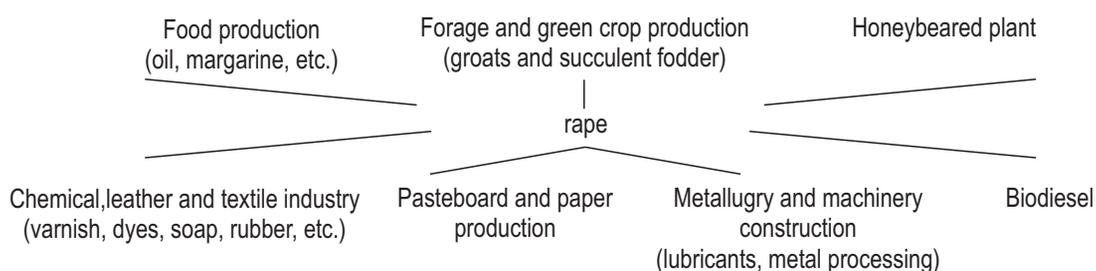


Figure 1. Different uses of rapeseed.

Table 1. Sown area and production

Years	Sown area, ha	Cultivated plots, ha	Average yield, kg/ha	Production, thousand tons
2006/2007	54 709	53 999	1723	93 018
2007/2008	93 730	87 804	2660	233 473

resistance to oxidation, both in the process of eating and under intensive heating (Staneva et al., 2007).

Two types of rape are grown - winter and spring. Rapeseed can grow at low temperatures. In most Northern countries, such as Canada, OSR is grown as a spring crop, while in other regions, e.g. in Europe (where winters are less severe), it is largely a winter-planted crop (Marjanović-Jeromela et al., 2008).

Discussion

Breeding

The first technical oilseed crop in Bulgaria in 1880 was rapeseed. During the war years the introduction of sunflower was

accelerated due to a shortage of oil. After 1965 the cultivation of rape ceased. It was replaced with sunflower because sunflower has significantly higher oil quality without low-quality ingredients, such as erucic acid in rapeseed oil and colza (Staneva et al., 2007). Rapeseed oil contains a large amount (45 %) of the harmful erucic acid (22:1) and glucosinolates which reduce its taste qualities. Genotypes with high erucic acid are suitable for biodiesel production. The Canon type is used in the food industry. This type has low erucic acid and glucosinolates. Erucic acid is a fatty acid that is not completely metabolized in the human organism and may cause deposition of fat in muscles and myocardial injury. Glucosinolates are protein compounds which are derivatives of amino acids and are decomposed in the animal body by the enzyme mirozinaza. In 1968 the first variety 'Oro' with oil free of erucic acid

Table 2. Fatty-acid composition in original and "OO" type of rapeseed (Gaivoronski and Balan, 2006).

Fatty acid / Glucosinolates	Quantity, %			
	Original rape*	Rape, type "OO"	Sunflower	Soybean
	Fatty acid			
C16:0 - palmitinic	3.8	2.5-6	4-6	6-10
C16:1	-	0.05-0.5	0.05-0.3	-
C18:0 - stearinic	1.1	0.7-2.5	2.6-6	2-5
C18:1 - oleic	11.2	45-65	14-38	20-30
C18:2 - linoleic	13.7	12-28	49-78	50-60
C18:3 - linolenic	8.1	4-10	0.05-0.27	5-11
C20:0	-	0.1-1.1	0.1-0.48	<0.01
C20:1 - eicosenic	9.6	1.71-2.5	0.05-0.29	<0.01
C22:0	-	0.04-0.5	0.3-1.38	<0.01
C22:1 - erucic	52.3	0.04-0.2	0.05-0.3	<0.01
	Glucosinolates*			
Gluconapine	33.3	5.5	-	-
Glucobrassicinapine	8.2	1.0	-	-
Progoitrine	109.4	8.3	-	-
Napoleipherine	5.2	0.4	-	-
Total:	156.2	15.3	-	-

*Raps GbR Saatzzucht Lundsgaard, 2010

was developed. The seeds of modern varieties contain 40-52 % of low-drying oil, 20 % of protein and over 17 % of carbohydrates. Gaivoronski and Balan (2006) have provided an example of the fatty-acid composition of the new "OO" rapeseed type as compared to sunflower and soybean (Table 2). New low eruc acid ("0" type) and low-glucozinolates ("00" type) varieties have been developed and the next stage in breeding will be a "000" type variety with low eruc acid, low-glucozinolates, and yellow seeds.

There are over 20 winter and spring rapeseed varieties included in the varietal list Bulgaria (Table 3). The standard varieties and hybrids for testing of biological and economic qualities (BEQ) are the protein varieties "Ural" and "Lara", as well as two winter forms: variety "Rasmus" and hybrid "Elvis". The first wintering Bulgarian oilseed rape variety "Marinus" ("O" type) was introduced in 1986-1987 by the Institute of Introduction and Plant Genetic Resources "Malkov", Sadovo. Two years later the first "00" type of winter oilseed rape variety "Yantar" was registered. The genotype of the progeny from the linear variety is homozygous unlike that of a hybrid (heterozygous). The method of propagation in the linear variety is

sexual self-pollination, while propagation in hybrids is by sexual artificial pollination. A hybrid variety is the result from crossing two genetically different parent forms (male sterile line and fertility restorer line) to enhance the vigour, viability and the production potential in F₁ (fertile), i.e. occurrence of heterosis effect. In late-maturing varieties it is better to use a hybrid.

Brassica napus L. is an annual (propagated through seeds) cross-pollinating plant with bright yellow flowers, which can also self-pollinate up to 70 %. Foreign fertilization (30 %) is possible by pollinators such as bees. Mature pollen grains are dual, with vegetative and generative nucleus. After sprouting the grain generative nucleus divides and forms two sperms. The pollen tube reaches the embryonic sac for 20-30 minutes, and the fusion of gametes and embryo formation occurs within 2-3 hours. The ovum retains its ability for pollination for a period of 4-7 days from the moment of flower opening. Pollen viability is high; it can last for a year under sterile conditions and low temperature, and may decrease under stress such as drought, high temperature and freezing. Often this leads to the occurrence of apomixis - the ovum is

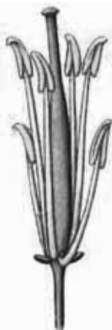
Table 3. Rape varieties included in the Official Varietal List of Bulgaria

Material	Type	Firm	Year of recognition
Spring			
Bufalo	variety	Norddeutsche Pflanzenzucht Hans-Georg Lembke KG, DE	2002
Ural	variety		2002
Lara	variety		2002
Eura	hybrid		2005
Jura	hybrid	Euralis Semences, FR	2005
Hunter	variety	Raps GbR Saatzucht Lundsgaard, DE	2005
Winter			
Panter	variety	173 „Canola EOOD”, BG	2002
Elvis	hybrid	Euralis Semences, FR	2002
Elite	hybrid		2003
Embleme	hybrid		2003
ES Artist	hybrid		2006
ES Astrid	variety		2006
ES Betty	hybrid		2006
ES Saphir	hybrid		2006
Olifant	hybrid		2006
ES Alias	hybrid		2007
ES Neptune	hybrid		2009
Rasmus	variety	Norddeutsche Pflanzenzucht Hans-Georg Lembke	2003
Captain	hybrid	KG, DE	2005
Rohan	hybrid	RAPS GbR, DE	2009
Vectra	hybrid	KWS SAAT, AG	2006
Triangle	hybrid		2006
Milena	variety		2006
Remy	variety		2006
Tassilo	hybrid	Limagrain Genetics G.C. S.A., FR	2008
Appolon	hybrid	Deutsche Saatveredelung AG, DE	2009
Hornet	hybrid	Pioneer Hi-Bred International Inc., US	2009
PR46W09	hybrid		2009
PR46W31	hybrid		2009

*Raps GbR Saatzucht Lundsgaard, 2010

already mature while pollen is still in state of gametogenesis (Poddubnaja-Arnoldi, 1976). Flowering begins early in the morning from the bottom of the flower and continues throughout the day, especially in wet weather (Musil, 1950). When the flower opens, the "muzzle" of stigma emerges first. The corolline petals elongate and the stigma is once again inside the flower – at the level of the stamens or lower. In most rapeseed varieties selective crossing occurs due to the different growth speed of the anthers. During pollination, it is necessary to pay attention to the simultaneous flowering of the varieties since there are great variations according to this index.

The generative organs of the rapeseed plant develop directly on the main stem. It branches off to varying degrees. A well-developed branch has its own flowers. Sepals are elliptical, 0.6 - 0.8 cm long and 0.15 to 0.2 cm wide. The four petals are light to dark yellow and almost twice as long as the sepals. Stamens have a characteristic red spot on the top. The two external stamens (out of six altogether) are shorter and with two nectaries. One plant forms 20-40 to 90 flowers. The duration of the whole plant flowering is 3 to 5 weeks. Low temperatures retard flowering and high temperatures accelerate it.



A cool and humid climate can make pollination difficult and cause formation of lower number of seeds per pod. Winter rapeseed needs about 60 days from beginning of flowering to maturation. During the flowering of plants, bee hives are placed near the crop (2 hives per 0.1 ha) to facilitate pollination. The pods on the main stem mature earlier than the pods on the branches. The fruit (pod) is narrow, 6-12 cm long and 0.35-0.45 cm wide. It could be diverted from the axis of attachment, or just hanging, oblate, with slight veining on the surface. The tip ends with a short beak. It consists of 2 bean valves separated by a false wall, which breaks at maturity. A single plant forms from 200 to 450 pods and individual plants can form up to 1000 pods. Each pod contains from 25 to 35 seeds, which are very small (diameter 0.15 to 0.3 cm) and spherical. Seeds are dark red to almost black, smooth or with slight veining. Recently, yellow-seeded forms are predominantly grown. Thousand seed weight varies within 3-7 g.

The stem of the rapeseed plant is cylindrical, branched, green, dark green or gray-green, with wax coating. Plant height varies from 50 to 200 cm, but most often it is from 80 to 150 cm. The stem is 1-5 cm thick at the base. The branching of the stem depends on the density of the crop, and first-order branches are between 12 and 25. The leaves are polymorphic. Rosette leaves are serrate, with handles, medium and top sheets are seated, serrate, heart-on and cover 1/2 or 2/3 of the stem. They are covered with wax coating and are blue-green in color. Their number can vary around 40. The vegetation period ranges from 280 to 320 days, of which 100 to 130 at low temperatures. Winter rapeseed varieties are divided into late-maturing, with vegetation period over 310 days, middle-maturing (280 - 310 days), and early-maturing - up to 280 days.

Rapeseed goes through the following phenophases: germination, sprouting, rosette formation, button formation, flowering and maturation. At optimum moisture and soil temperature (14-15° C) the seeds germinate for 3-6 days, and the first true leaf emerges in 10-15 days. One month after germination, plants form a rosette of 6-8 leaves which reaches 30-60 cm in diameter at the end of vegetation. In this stage winter rape survives through the winter season. The high cold resistance of rapeseed is determined by the large amounts of sugars accumulated in the roots, the root collar and the points of growth. Spring growth is resumed after the mean daily

temperature increases over 1-2°C, and soil temperature - over 2-3°C for 9-10 days. Two weeks after the resumed spring growth the formation of stem begins. Button formation stage begins 10-20 days after the beginning of spring vegetation. The period from button formation till flowering is 20-25 days. Three to five flowers open per day and 40-55 % of the flowers bear pods. The number of flowers depends on the varietal characteristics and the climatic conditions. Under controlled conditions, 68 % of the flowers develop into pods and the rest fall off. The higher temperature during the vegetative development in comparison to the temperatures during flowering and pod filling leads to higher yields. When flowering occurs at lower temperatures, higher yields are obtained. Maturation of winter rape follows the course of flowering, i.e. from base to tip, and the seeds mature for 35-45 days after flowering.

The traits for screening are higher oil content in seeds and the absence of erucic acid in it for varieties used in the food-processing industry, as well as high oleic (70 %) and low linolenic acid (up to 25 %), high yield, high content of oil and protein in seed, uniform development, flowering and maturation, earliness, resistance to lodging, pod dehiscence, high yields of green biomass, resistance to diseases and pests, high winter and frost resistance and seed coloration. Recently yellow seed varieties are preferred due to their high oil and protein content. Yellow seed with a thin coat is of higher quality than in the dark-colored seeds. Technical oils (hydraulic and lubricating) and biodiesel need high erucic acid content; the oil for the perfumery industry and for the production of synthetic materials needs high content of lauric acid.

The initial phase of work on rapeseed is the selection of landraces and foreign accessions and populations. Mass selection by phenotype till flowering of the plants is the easiest way, followed by mixed seed sowing of individual plants. This method is not labour-consuming, although working with large populations, but it does not allow controlling the selected genes. Recurrent selection is used to alter the fatty-acid content in rapeseed, which allows gradually increasing the frequency of genes which control the breeding trait. Individual and family selection is also used, which could be combined with inbreeding. In rapeseed, breeding methods such as intraspecific (intervarietal and pedigree) and interspecific hybridization have been used (rapeseed has been crossed to cabbage, mustard and other species of genus *Brassica*) (Prakash and Hinata, 1980; Warwick and Black, 1993; Scheffler and Dale, 1994; Brown et al., 1997; Bajaj et al., 2004, etc.), as well as backcrossing, mutagenesis (chemical and gamma irradiation), re-synthesis, haploidy (a frequently manifested spontaneous phenomenon - apomyxis, use of colchicine to increase the chromosome number). The haploids possess smaller flowers that have no fertile pollen. The methods for production of haploids include cultivation of anthers, pollen and microspores. Since *B. napus* is tetraploid, it will cross more readily with wild species (diploid) as a female parent (Sikka, 1940; Harberd and McArthur, 1980). In the case of *Raphanus raphanistrum*, no difference was noted in the direction of crosses (Kerlan et al., 1991), in the case of *Sinapis alba*, the opposite situation occurs (Ripley and Arnison, 1990).

In genus *Brassica*, there are six types of chromosomes ($n = 6$, pahiten analysis) designated by the letters *A, B, C, D, E* and *F*. In most species of this genus chromosome types are found, which repeat with a different number. Thus for example, the initial forms of genus *B. nigra* ($n = 8$) have chromosome composition *ABCDDEEF*, *B. oleracea* ($n = 9$) - *ABBCCDEEF* and the chromosome composition of *B. campestris* is *AABCCDEFFF* ($n = 10$) (Munz, 1968; Muenscher, 1980). Interesting is the origin of rapeseed since there is no wild form

of this plant. Rapeseed resulted from crossing *B. campestris* (2n = 20, genome AA) to *B. oleracea* (2n=18, genome CC) and the subsequent doubling of the chromosome number – an instance of natural amphidiploidy. Rapeseed (*Brassica napus oleifera* D.C.) has a spring (*B. napus oleifera annua* Metzg.) and a winter (*B. napus oleifera biennis* Metzg.) form. The karyotype is with 38 chromosomes (n = 19, genome AACS) combined in the germ cell in the following way: AAABBBCCDDDEEEFFFF.

Most of the rapeseed traits (such as stem height, number and size of leaves, number of branches, rosette size, length of growing season, winter and drought tolerance, protein content in green mass, etc.) are inherited intermediately in F₁, but in F₂ transgressions and additive effects occur. There is a negative correlation between oil and protein contents in seed. Anthocyanin coloration on the petiole and stem, and the presence of wax coating are dominant. Erucic acid is controlled by five alleles with additive effect (*e, Ef, Eb, Ee, Ed*). There is also a correlation with the direction of crossing - the erucic acid content is higher if the female parent is a high-erucic variety. In F₂ generation the highest number is that of erucic-free and low-erucic plants in the combinations in which the female plant is low-erucic. The erucic-free varieties are less productive. In the inheritance of glucosinolates the role of cytoplasm is important. Their level increases with the increase of 1000 seed weight, but it is inherited regardless of the trait erucic acid content. The synthesis of palmitic and stearic acid is controlled by a poligenous system with multiple alleles at the loci.

In recent years the breeding programmes focus on development of hybrids. To achieve this aim it is necessary to find out new CMS sources and fertility restorer genes (*Rf* genes) and transfer them into genotypes which are carriers of genes responsible for agronomically important traits. The male sterility and fertility restoration system is used in most open-pollinated crop species when positive effects of heterosis for agronomically important traits need to be exploited. Male sterility, although generally defined as the condition where viable pollen is not produced, is variable in expression and can range from complete absence of stamens to their dehiscence and release of normal viable pollen (Vipen and Shukla, 1994). In most cases, CMS trait is a result of interspecific or intergeneric hybridization (aloplasmtic sterility). In rapeseed, the Ogura-CMS system (INRA - France) and the MSL-system (German Lembeke Institute) are most commonly used. Ogura / INRA CMS system was obtained using protoplast fusion between rapeseed and radish (*Raphanus sativus*). MSL system was obtained thanks to a spontaneous mutation and selection during backcrossing. The CMS trait is unstable, because partial fertility is possible under high temperatures. The Ogura CMS system, unlike others, is stable under various climatic conditions. The main problem for Ogura CMS is its fertility restoring gene (*Rf* gene), which is linked tightly to the genes that determine high glucosinonates content (Renard et al., 1997, as referred to by Yang et al., 1999). Atlagić et al. (2008) using cytogenetic methods analyze the stability of CMS after introduction into rapeseed inbred lines and identify the differences between various CMS types. All of the inbred lines analyzed were male sterile with different levels of anther development. The backcross progenies showed that both CMS sources are stable, and restorers were found for two genotypes. The analyzed types of CMS all had normal meiotic phases, but the post-meiotic division showed irregularities that led to the interruption of microsporogenesis, most frequently at the phase of tetrads. Similar results were reported by Atlagić et al. (2003) and Atlagić et al. (2007).

Significant work has been done in the breeding for resistance to

various diseases and pests. Using genetic engineering, the oxalate oxidase (OXO) gene was transferred from wheat to rapeseed. It reduces the symptoms of *Sclerotinia* but GM canola should be studied to find out exactly how the climate changes affect this gene (Lu, 2000; Scheffler and Dale, 2005; Dong et al., 2008, etc.). GM rapeseed has been grown in Canada since 1996. In 2003, GM oilseed rape occupied 3.6 million ha, and in 2006 - 4.8 million ha (20 % of sown areas). In 2007, GM rapeseed was grown on 5.1 million ha, which were approximately 87 % of Canada's rapeseed crop. GM rapeseed is grown on smaller areas in the US and in certain states in Australia (GMO Compass, 2010). Table 4 presents the new traits. Admittedly, the biotechnology methods are able to transfer genes from other plant families, even viruses, bacteria, fungi and animals. For them there are no barriers of incompatibility.

Table 4. Traits in GM rapeseed cultivated and/or consumed worldwide.

Desired effect (enzyme, gene or factor)	Region
Glyfozitol tolerance (EPSPS, EPSPS + GOX)	Australia, ES, USA
Glyfozinatamonii tolerance (PAT)	Australia, ES, USA
Bromoxynil tolerance (Nitrilase)	Australia, USA
Male sterility (Barnase); Fertility restorer (Barstar)	Australia, ES, USA
Lowering of phytase	USA
High laurea content in oil	USA

Last year Dobrudzha Agricultural Institute (Bulgaria) started a new breeding programme for development of Bulgarian varieties which could meet the new agronomic and economic conditions. The first step is the collection and characterization of initial material. The potential of introduced winter and spring rapeseed varieties is investigated. The technology for growing new varieties and hybrids developed in Europe and worldwide is studied and improved. Work is directed towards developing of parental forms with transferred genes for economically important traits such as presence of *Rf* genes, genes for control of resistance to diseases and pests, and use of the heterosis effect.

Cultivation, sowing and harvesting

There are critical stages in the cultivation of winter rapeseed during each season. Autumn is the most important time for germination and during the winter there is the danger of frost. In spring it is time for integrated pest control practices. During this season there may be conditions for lodging as well. In summer diseases such as *Sclerotinia* occur, additionally rapeseed is under drought and high temperatures. Another negative factor may be the presence of excessive moisture during harvesting.

The soil and climatic conditions in Bulgaria are suitable for growing canola. It can be grown on all soil types with a neutral pH reaction. Most favourable are the rich fertile soils, as "chernozem" and dark-gray forest soils with clay-sandy mechanical structure. Unfavourable soils are inadequate light, sandy, marsh and saline soils. An important condition is that soils do not form crust. Rapeseed requires temperate climate. It is a plant of the long day. Shading

leads to reduction in the number of flowers and the number and size of pods and seeds in them. The shortage of light during seed filling is the main reason for seed weight decrease by 15-20 %. In order to germinate, seeds need 70 to 100 % water according to their weight. In case of insufficient moisture in the surface layer during sowing seeds remain in the soil for a long time without germinating. For germination and initial development of rapeseed, 20 mm productive moisture is needed in the surface layer. The greatest requirements of canola are in stages flowering, button formation and seed-filling. To ensure the necessary quantity of vegetation moisture, rainfalls of 45-50 cm are necessary. During flowering rapeseed is also sensitive to atmospheric humidity. Precipitation and low temperature during the course of flowering reduce the number of flowers, and the number and size of pods and seeds in them. Warm and dry winds during this period will reflect negatively on pod formation. As a result the period of flowering is sharply shortened and large numbers of flowers remain unfertilized, the formed seeds are shriveled and malnourished, which significantly reduces the amount of seed yield. This is the reason for the lower yields obtained from the spring varieties. A major problem of growing canola in Bulgaria is the shortage of water. Therefore, very dry autumns and springs require two irrigations - one for germination and another during flowering. Since spring is usually wet, most often it is enough to do only the first irrigation.

In winter, rapeseed is able to survive under very low temperatures. A prerequisite for its successful cultivation is its tolerance to low temperatures. The best time for hardening of the plants is during "rossette" stage at temperature 5°C for 10 days, and at -3°C for 5 days. Well developed and hardened plants can survive low temperatures up to -15°C without snow; when the snow cover is more than 5 cm, the plants can survive under -25/-30°C. Non-hardened plants die at 6-8°C. Rape is hypersensitive until stage 4th leaf. In the cool and humid weather of early October, the well developed rapeseed plants continue to grow and those at stage 2nd leaf cease their development. The plants damaged by frost grow slower during the spring. Critical to the development of rapeseed are late spring and early autumn frosts. Temperature which causes damage depends on the degree of development of plants, the level of moisture and the length of freezing temperatures. Highly wet soil and frequent alternation of warming and frost cause severe damage to the crop. While in dry soils plants die at temperature below -16°C, in soils with high moisture damages by frost are observed even at -7-8°C. The most favorable temperature for growth and development of canola is 18-20°C. Air temperature of up to 23°C is needed during flowering and ripening.

In the cultivation of rape there is a danger of loss of plants during dry and cold weather when the crops are not covered with snow or the snow cover is insufficient to protect plants. In winter days with good sunshine, rape begins to photosynthesize, taking up and evaporating moisture in the process. At the same time, however, soil temperatures are below normal and the plants cannot absorb water and they perish because of cold and drought (physiological drought). Another problem is the thick snow. Then the concentration of carbon dioxide from the snow increases and plants suffocate. Temperature variations and frequent freezing of soil can lead to root death.

Rapeseed is characterized by its powerful root system. Under favorable soil conditions this oilseed crop develops a highly branched taproot (axial type) with deep roots at stage first pair of leaves, and the root system reaches 30-40 cm in depth; in one month (4th -5th pair of leaves) the root is 60-70 cm deep, and 150-180 cm at the end of button formation; at maturation it reaches 300 cm,

but the main mass of roots is concentrated in the 60-90 cm soil layer. Rapeseed has a deep root system which is preferable to the shallow root in cases of drought. Taproots may not fully develop when there is soil compaction, in the presence of undecomposed organic substances or when there are damages by soil acidity. This may affect cold resistance, regeneration capacity and yield. Before winter rapeseed must have a sufficiently developed root system - taproot length of 20 cm, and be at least 0.5 cm in diameter to be able to use deeper soil moisture.

For the good cultivation of rapeseed it is necessary to provide good conditions for germination and development of young plants - timely soil tillage, fertilization, sowing and adequate control of weeds and pests. Furthermore, winter conditions, soil cultivation and sowing influence the achievement of high and stable yield. Crop rotation should be at least 3-4 years to avoid disease attacks. The most suitable previous crops are cereals and some legumes (although they are sometimes attacked by the same diseases). Unsuitable predecessors are sunflower (both crops are hosts of many economically important diseases) and beet (increased risk of nematodes). The best previous crop is wheat. Cereals are removed from field earlier (by mid-August), leaving soil with good moisture reserves and free of weeds. Investigations carried out in Germany revealed that oil content in seeds of winter oilseed rape grown after winter wheat crop is 0.9 % higher than the rapeseed grown after wheat-clover or clover mixes (Staneva and al., 2007). Winter oilseed rape is sown after winter wheat, grain-legumes, and root crops. Rapeseed should not be sown after cabbage species.

Germination and initial development of seeds depend on the cultivation of soil (soil tillage should be excellent). Thousand seed weight should be 4.5 - 5 g, and the germination capacity of seeds - over 96 %. Higher sowing norms at low germination are not recommended, since plants with reduced vigor develop. Certified seeds should be used for sowing, which are large, plump, cleaned and produced in normally developed seed crops. When choosing a variety of rapeseed, the following should be considered: 1. conditions of the region - infection of the sowing areas with weeds, weed composition; 2. conditions for development of diseases and pests; 3. expected rainfalls and their distribution during the growing season; 4. amounts and temperature amplitudes. Cultivation depth varies between 20 and 25 cm so that soil can accumulate the necessary amounts of precipitation in autumn. Cultivation has to start immediately after harvesting the predecessor to preserve the remaining moisture in the soil, especially during dry summers. Straw must be left on the surface and fixed with soil so that it can be incorporated in deep tillage (scuffed stubble). Harrowing and disking should be applied once or twice according to the condition of the field. Soil should not be allowed to dry because large lumps are formed and soil compaction may be a problem. Before sowing of rapeseed, harrowing and packing of soil should be done to form a smooth surface and a solid bed for seeds. Solid bed allows capillary rise of water to seed, which in turn helps them germinate.

Rapeseed is sown in late August - early September (the most suitable period is 25.08-15.09). The sowing dates must be observed precisely, depending on the weather during the crop year and with a view of the plants forming in autumn a rosette of 6-8 leaves in order to successfully survive in winter. Shallow sowing (up to 2 cm deep in dry conditions and up to 4 cm in normal ones) should be performed directly after soil tillage to allow the seeds to better use soil moisture. The choice of a variety or hybrid is important, depending on the region. On plain areas varieties can be grown, but on the more difficult hilly areas where severe winters are expected hybrids are recommended for sowing. The quantity of seed for planting depends

on the type of cultivar - linear variety or hybrid type. Full details should be written on the labels attached to the bags of certified seeds. Germination capacity of 95 % is considered good. For linear varieties the quantity of seeds is as follows: 50 germinating seeds per m² for early seed (until August); up to 50-65 germinating seeds per m² for normal seeds (1st-10th September); up to 70-90 germinating seeds per m² for late seed (10th September). In hybrid varieties the quantity of seeds should be the following: 40 germinating seeds per m² for early varieties; up to 40-55 germinating seeds per m² for varieties with an intermediate vegetation period; 65 germinated seeds per m² for late varieties. The optimal density is between 45-50 plants per m² for hybrids and 60-70 plants per m² for varieties. The distance between rows should be 25 cm (12-15 cm at sowing norm 4 – 8 kg/ha for production of seeds and 10-12 kg/ha for production of green mass to provide a minimum of 20-30 plants/m² for the

harvesting). Higher sowing norms cause reduction in the number of branches and pods, and thus lower yields are obtained. Dense crops are prone to lodging, since plants are higher.

Fertilization is done according to the necessary amounts of nutrients (balanced nutrition), having in mind that rapeseed is nitrogen-dependent crop and takes up nitrogen from soil in amounts two times higher than cereals because it needs high quantities of nutrients during the first stages of its development (September-November). The nitrogen norm is 60-130 kg/ha up to 170 kg/ha; 40-60 kg of this norm are applied in the autumn with the pre-sowing tillage of soil, together with phosphorus (50-80 kg/ha) and potassium (50-100 kg/ha), and the rest is applied in spring. Excessive nitrogen fertilization leads to excessive green vegetation, and increases the risk of partial or total freezing of plants. The shortage of nutrients (potassium, calcium, magnesium, boron) is manifested by different symptoms – chlorosis developing into necrosis of leaves at different

Table5. Primary treatment.

Treatment against	Pesticides	Time for treatment
Annual cereal and broad-leaved weeds	Agriflan 24EK, Butizan 400SK, Butizan S, Devrinol 4F, Sultan 50SK, Triflusan 48EK	October- before sowing, incorporation
Annual and perennial cereal weeds	Galant Super, Targa Super, Fuzilad Super	November
Wild mustard, turnip, charlock	Derby 175SK, Sansak	
Corn-thistle, camomile, mayweed	Lontrel 300EK	
<i>Athalia colibri</i> (ETD is 2-3 maggots per 10 damaged plants per m ²), <i>Entomoscelis adonidis</i>	Bancol 50VP, Dursban 4E, Karate 5EK, Marshale 25EK, Mospilan 20SP, Nurele D, Pilot 48EK, Tombel 32EK, Vaztac 10EK	November
<i>Ceutorhynchus napi</i> , <i>Meligethes aeneus</i> (ETD is 1-2 pollen beetles per plant and 6-8 pollen beetles per plant at stage "button formation"), <i>Brevicoryne brassicea</i> , <i>Psylliodes chrysocephala</i> (ETD is 2-4 numbers per m ²), bugs and etc.	Bancol 50VP, Vaztac 10EK, Marshale 25EK, Mospilan 20SP, Nurele D, Lambda-cyhalothrine and etc	During the period from March-April till June
<i>Sclerotinia</i> , <i>Phoma</i> , <i>Cylindrosporium</i> , <i>Oidium</i>	Flutriafol + Carbendazime, Flusilazole + Carbendazime; Topsin M 70VP	March-April
Powdery mildew	Alto 320EK, Panc 40EK, Sistan Super 24E, Topsin M 70VP	April-May
Downy mildew, White rust, Black rot, Dry stem rot	Vitra 50VP, Ditan M45, Cupertin M	May-June
Gray rot	Rovral 50VP, Sumilex 50VP	
Leaf spots	Benomil 50VP	
Goitre	Solution with hydrated lime	
Weevil, wire worm and others soil pests	Furadan 35ST	Seed disinfection
Against pod dehiscence and for diminishing losses before and during harvesting	Elastic TM	Before harvest

levels (lack of potassium and magnesium), growth stunt, shortening of internodes, strong lateral branching of stems, flower formation is stunted, decrease of pod formation with 20 % (boron insufficiency), epinasty - droop of the flowering tops of peduncles, followed by necrosis (calcium insufficiency). Introduction of boron in the soil can be done by seed treatment or by leaf fertilization. The optimal norm of fertilization is 0.3-0.5 kg/ha. Boron may be applied in the form of borax, boric acid, superphosphate, or ammonium nitrate with higher content of boron. Its introduction leads to increased winter-resistance, prevents lengthening of plants, helps the formation of generative organs. Formation of high yields and resistance to late-spring frost requires sulphur. The ratio between nitrogen and sulphur should be 15:1. When there is sulphur deficiency, the young leaves are reddish, and the corolline leaves are pale yellow to white. Growth processes are stunted and the plant becomes chlorotic; yield decreases with 10 %. Sulphur deficiency is most often observed after great temperature variations. Highest effect on light and moderately heavy soils is observed after application of 30-40 kg/ha sulphur from the beginning of the growth of stems (over 20 cm) till flowering. Heavier soils need fertilization with 20-30 kg/ha sulphur. Fertilization with molybdenum has good effect on grey forest soils, when rapeseed has been already treated with higher nitrogen norms. Attention should be paid to the fact that for the formation of 100 kg of seed per hectare and the respective shoot, rapeseed takes up from soil the following nutrients: 50-60 kg/ha N; 20-30 kg/ha P₂O₅; 40-60 kg/ha K₂O; 40-70 kg/ha CaO; 40 kg/ha sulphur.

Diseases and pests can be serious problem in the cultivation of rapeseed. Rapeseed is suffering from both non-infectious and a number of infectious diseases. These are the more important diseases on this crop: downy mildew, powdery mildew, leaf spots, stem dry rot, white rot, gray mould, black rot, *Cylindrosporium*, *Sclerotinia sp.*, *Phoma sp.*, white rust and etc. (Boyles et al., 2006; Foubert, 2010). Rape is attacked by many pests (several aphid species, rape bug, rape weevil, etc.) which, when reaching certain density, can cause extensive economic losses and even compromise the entire crop. All plant parts are subjected to attack - roots, leaves, flowers, pods. Therefore the use of appropriate plant protection products is mandatory when the economic threshold of damage (ETD) is exceeded. The care for the crops during the growing season is limited to maintaining the areas free of weeds, spring fertilization with nitrogen, irrigation and disease and pest control (Table 5).

Weed control begins with soil treatment with herbicides. The addition of fungicide substances and seed treatment with insecticides are very important. These substances protect the embryos and the young plantlets allowing the development of symmetrical plants with a sufficient number of plants per m². Pest control is done by appropriate agronomy practices and by chemicals, correct crop rotation, control of cruciferous weeds, isolation of other cruciferous crops, spraying with insecticides during the growth period. Spring pest control is directed towards cruciferous fleas, various types of cabbage bugs, caterpillars of cabbage butterflies and others. Most difficult is pest control during flowering and maturity because the pollinators, especially honey bees, have to be protected. Therefore during this period it is best to use pyrethroids. Disease-tolerant varieties and hybrids are recommended.

Harvesting is done in June-July with harvester, this is direct harvesting at stage full maturity of seeds (brown and hard, with moisture content up to 15 %), before pod dehiscence (higher pod dehiscence means greater seed losses). The most favorable time is

in the early hours of the day or late in the evening. It is necessary to know the characteristics of the respective variety with regard to unfavorable conditions such as lodging, earlier pod dehiscence, etc. The critical moisture content of seeds of the different varieties is from 8.3 to 8.6 %, but in longer storage humidity should be 7 % because the seed have high oil content, which, as in all oilseed crops very easily oxidates.

The yield is determined by the number of plants (per area and per plant) and by 1000 seed weight. For maximum yields, rapeseed should have leaf area index and pod surface indexes from 3 to 4:1.

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