BREEDING AND SOME BIOLOGICAL PROPERTIES OF YELLOW SEEDED WINTER RAPESEED (Brassica napus L.)

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The quality of rapeseed meal as an important source of protein is limited by its high content of crude fiber in the seedcoat. In studies on different Brassica species (BECHYNE et al. 1979, BELL and SHIRES 1982, LIU-HOU-LI et al. 1983, STRINGAM et al. 1974, SHIRZADEGAN and RÖBBELEN 1985) have shown that yellow seeded forms have got a considerably thinner seed coat than dark ones, contain a smaller proportion of hulls, resulting in improvements in oil, protein and fiber content. At present all the cultivars of rapeseed (B. napus) licensed in Europe are dark in seed color.

Interspecific hybridization within the genus Brassica has been well recognized as an important tool for transfer the yellow seed coat color, from B. campestris or some other species to B. napus (BECHYNE 1985, 1986). The transfer of seed coat color, seed testa of interspecific hybrids offers an interesting field of study. This is the object of our experiments in this paper which presents some results of yellow seeded B. napus.breeding in Czechoslovakia.

MATERIALS AND METHODS

Materials utilized in interspecific crossing for obtaining yellow seeded B. napus originated from Brassica campestris - var. Yellow Sarson, B. oleracea var. acephalla (selected light greyish seed) and yellow seeded B. carinata (selection from a land race). Plants with crossed buds were kept in isolation chamber till the stigma was seen to be developing into a pod. Progenies were raised in pots, greenhouse beds and advanced generations were field grown.

Four classes of color for the main analyses and six for microscopic studies were identifiable and the seed was classified as either yellow, ochre, brown and black in the first case and yellow (I.) ochre (II.) yellow brown (III.) light and dark brown (IV., V.) and black (VI.). For microscopic measurements of testa, seeds were dehydrated and saturated thoroughly directly in ethanol - n-butanol mixture (15 : 85) for 12 - 24 hrs. and then saturated with dehydrogenated pure n-butanol and slowly transfered into paraffin. The sections were stained in Ehrlich hematoxylin and enclosed into Solacryl resin.[x)] Considering the expected sterility of some hybrids, colchicin treatment was made on seedlings at 2-4 leaf stage. 0,01 colchicine solution was applied by cotton pads to the vegetative tips, five times a day. The oil content of the whole seeds was measured by NMR spectroscopy. For further analyses and measurement conventional techniques were used.

RESULTS AND DISCUSSION

Interspecific hybrids between B. campestris and B. carinata (and reciprocals) set sufficient number of yellowish-grey hybrid seed. 162 crosses and reciprocal crosses formed 17 pods with 65 seeds (0,40 seed per pollination). The seeds developed into F_1 B. carinata like plants with vigorous growth and disproportionaly long branches with sufficient number of pods mostly without seeds. A few pods contained some misshapen sied like structures, which did not germinate in favourable conditions on moist filter

x) Microbiologic studies were provided by J. Pazourek and staff, Faculty of Natural Science Charles University, Prague.

paper. The sterility barrier could not be overcome by chromosomal duplication. The results are not in agreement with MIZUSHIMA (1950), who reported 50 % fertility in allopolyploids of his B. campestris X B. carinata hybrids. But the pods and seed like structures indicate that a succesful fertilization in some cases is affected, but the embryo development is inhibited at some later stage. The embryoculture could probably overcome the sterility. The sterility barrier was the reason for not taking further full advantage of B. campestris x B. carinata crosses.
A total of 120 pollinations betwenn B. oleracea and B. campestris was made. One cross combination only proved to be successful, producing 0,18 seed per pollination. After doubling the chromosome number only one plant proved to be tetraploid and survived up to maturity. It showed many signs of intermediate character. One of the hybrid plants produced in F_2 generation yellow seeds. After repeated selfing and selecting the best shade of yellow color was the spring type of yellow seeded B. napus crossed with Jet Neuf. The hybrid plants formed after continuous selfing and backcrossing a winter form of yellow seeded B. napus. Its seed however was not bright and deep in yellow color as in B. campestris, carinata or juncea. The properties of the best yellow seeded plants were evaluated in four years field trials and the characters of seed coat different in color, obtained within the breeding were compared (as shown in tables and figures).

Table 1: The overwintering of B. napus plants in %.

	1982	1983	1984	1985	1986
Yellow seeded	64.4	91.6	86.4	89.6	83.1
Jet Neuf	87.5	96.9	92.7	91.0	84.3
Difference	23.1	5.3	6.3	2.4	1.2

The overwintering of yellow seeded plants, improving in successive generation (except for 1984) was significantly lower in 1982 - 1984, compared with Jef Neuf (Table 1).

The microscopic measurements show an evident improvement in yellow and light colored seedcoats. Yellow seeds are the longest in radius (917,9 μm), while the black seeds reached 849,3 μm only. The thinner testa, mainly due to the palisade layer, explains the differences in biological and biochemical properties of the yellow seeded material. The two measured testa layers appear to be highly positively correlated (parabolic course). The black seeded strains differ evidently from the light ones.

The means of four years field experiments (tab. 2) show that the yellow and light seeded strains do not fully reach the properties of Jet Neuf. (No. of primary branches, No. of pods, per plant and oil content.) But the seed coat thickness and crude fiber content evidently elevate the former seeds. However it should be pointed out a slight variation in seed color shade in yellow seeds. Even within single pods brown or yellow-brown spots, mostly around hilum can be seen, similarly as indicated by HEYN (1973), JÖNSSON (1975) and HAWK (1982). XIAO found out in 1982 that the lighter the color of seedcoat, the thinner the seedcoat and the higher the oil content. His results support our intentions and necessity for further breeding of hight yellow color, which could improve the economic properties of yellow seeded rape plants and overtake the present cultivars.

Table 2: Biological and economic characters of plants.

Character	Seed coat color			
	yellow	ochre	brown	black (Jet Neuf)
Height of plants (mm)	126.4	128.1	126.3	132.6
No. of primary branches	5.5a	5.7a	5.7a	6.2b
No. of pods per plant	120.6a	121.3a	124.0a	131.0b
No. of seeds per pod	15.6a	15.8a	15.6a	16.3a
1000 seed weight (g)	4.4a	4.3a	4.4a	4.5a
% of seed coat	11.6a	12.1a	17.0b	19.7c
Oil content %	44.8	44.9	45.3	45.4
Sum oil + protein %	67.8	67.1	65.1	65.3
Crude fiber %	10.2a	10.6a	12.7b	13.1b
Yield (t ha^{-1})	2.59b	2.52b	2.43a	2.77c

Means within rows followed by different letters are significantly different (PL 0.05)

SUMMARY

Winter forms of yellow seeded rapeseed (B. napus L.) were obtained as a cross of yellow seeded spring B. napus line and winter rapeseed cv. Jet Neuf. The yellow seeded spring rape originated from the interspecific crossing (B. oleracea and B. campestris). Four classes different in seed color for main analyses and six for microscopic studies were identified. The microscopic measurements of seed coat showed evident positive characters in yellow and light colored seeds (seed size, thinner testa layers). The differences in microscopic studies support the efforts for obtaining better seed properties (less fiber, more oil and protein and pleasing light appearance of meal), even if they do not reach fully the excellent parameters of Jet Neuf in our experiments. Further breeding and overcoming a slight variation in yellow seed color is necessary.

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Table 3: Statistical parameters of seedcoat characters in six different colors (I. - VI.)

Testa color	Seed radius (µm)			Testa thickness (µm)			Palisade l. thickness (µm)			Thickness of aleurone l. (µm)		
	$\bar{x}$	s	V_k	$\bar{x}$	s	V_k	$\bar{x}$	s	V_k	$\bar{x}$	s	V_k
I	917.90	17.534	1.91	36.80	4.770	12.96	14.66	1.728	11.79	22.14	3.856	17.42
II	882.00	19.241	2.18	43.66	6.091	13.95	17.58	1.798	10.23	26.08	4.590	17.60
III	913.95	19.271	2.11	39.63	4.295	10.84	16.78	1.174	6.99	22.85	3.431	15.02
IV	896.55	5.325	0.59	40.28	5.360	13.31	16.62	2.233	13.38	23.56	3.734	15.85
V	876.55	12.540	1.43	38.11	4.526	11.88	17.17	1.434	8.35	20.94	3.417	16.32
VI	849.30	7.390	0.87	60.32++	5.796	9.61	35.86++	3.199	8.92	24.46	4.091	16.73
	Ratio palisade-aleurone ls.			Thickness of palisade l. in testa (%)			Thickness of testa in seed radius (%)			Thickness of palisade l. in seed radius (%)		
	$\bar{x}$	s	V_k	$\bar{x}$	s	V_k	$\bar{x}$	s	V_k	$\bar{x}$	s	V_k
I	0.677	0.117	17.35	40.089	4.177	10.491	4.011	0.541	13.48	1.596	0.180	11.29
II	0.682	0.071	10.41	40.512	2.607	6.436	4.949	0.673	13.60	1.995	0.201	10.06
III	0.748	0.091	12.24	42.561	2.895	6.802	4.339	0.504	11.61	1.836	0.141	7.69
IV	0.714	0.097	13.66	41.418	3.578	8.639	4.491	0.585	13.03	1.854	0.242	13.03
V	0.832	0.098	11.82	45.279	2.927	6.465	4.348	0.522	12.01	1.995	0.155	7.94
VI	1.493++	0.231	15.45	59.58++	3.810	6.395	7.108++	0.702	9.88	4.223++	0.391	9.25

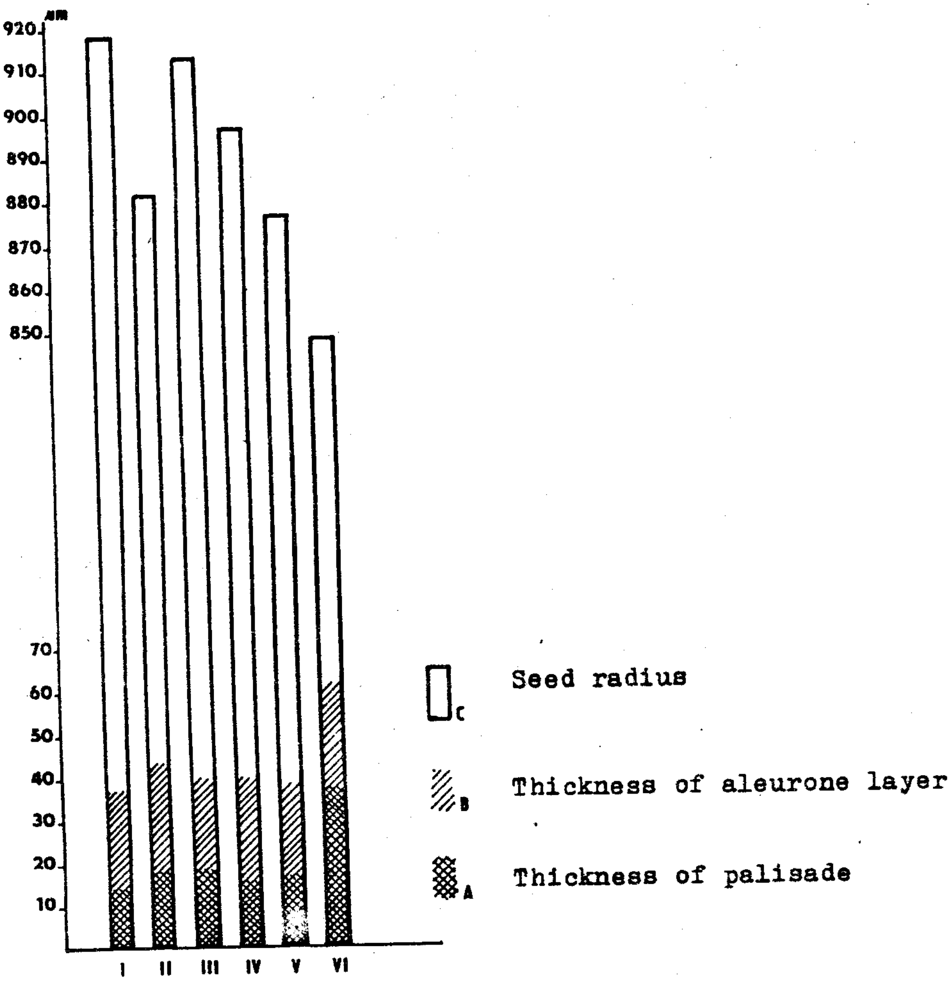
++ ... P=0.01

Table 4: Correlation index values - palisade and aleurone layers

Testa color	I	II	III	IV	V	VI
Plants 1	0.3538^{++}	0.2050	0.2849	0.1650	0.1778	0.5864
2	0.4460^{++}	0.1463	0.1654	0.3570	0.4578^{++}	0.5538^{++}
3	0.2727	0.5164^{++}	0.3250^{++}	0.4877^{++}	0.1628	0.4356^{++}
4	0.4107^{++}	0.6650^{++}	0.4380^{++}	0.1155	0.1140	0.2700
5	0.4969^{++}	0.5435^{++}	0.5011^{++}	0.5534^{++}	0.2822	0.2101
6	0.2057	0.5257^{++}	0.2703	0.1037	0.4160^{++}	0.5377^{++}
7	0.5594^{++}	0.2041	0.4949^{++}	0.2135	0.4029^{++}	0.1650
8	0.5608^{++}	0.1822	0.2979^{+}	-0.0898	0.2966^{+}	0.3216^{+}
9	0.1676	0.5803^{++}	0.2137	0.2579	-0.1434	0.3323
10	0.1308	0.5100^{++}	0.1048	0.3541	0.2497	0.4092^{+}
1-10	0.3172^{++}	0.4509^{++}	0.3077^{++}	0.3131^{++}	0.3128^{++}	0.2445^{++}

(+ ... P=0.05, ++ ... P=0.01)

Fig. 1 Thickness of seedcoat layers and seed radius.



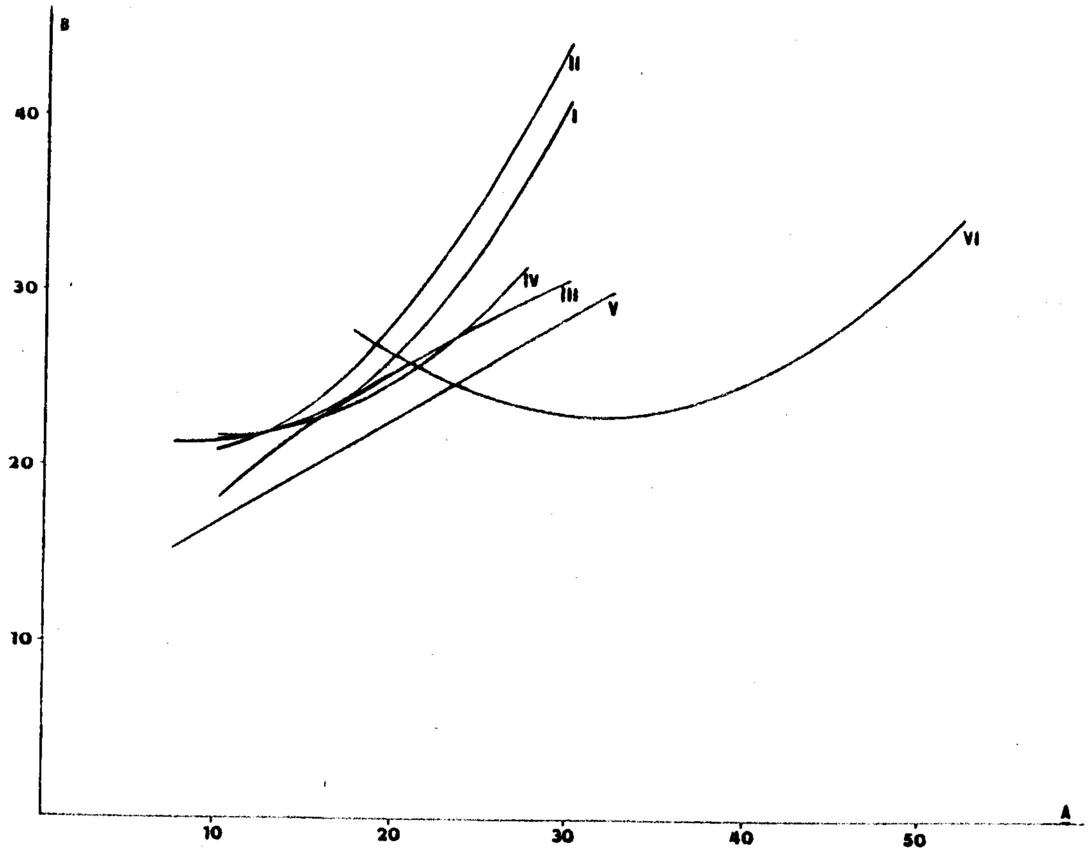


Fig. 2 Correlations between palisade (A) and aleurone layers (B) in six seed coats different in color (I. - VI.)